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HÖFCHEN-BRIEFE

BAYER PFLANZENSCHUTZ-NACHRICHTEN

English edition

No. 1, 1960

PUBLISHED BY THE BAYER CROP PROTECTION DEPARTMENT
LEVERKUSEN AND HÖFCHEN

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BAYER LEVERKUSEN (GERMANY)
Department for Crop Protection

Crop Protection Research Laboratory, Farbenfabriken Bayer Aktiengesellschaft, Wuppertal-Elberfeld Works

Development of the Bayer Product "S 1752"

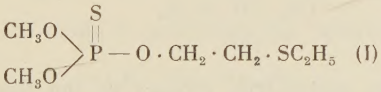
by G. Schrader

The aim of chemists carrying out research in the crop protection branch is to supply agriculturists and other users of pesticides with substances which a) have a low order of mammalian toxicity, b) feature a very wide spectrum of activity against pests, and c) have no harmful effects on beneficial insects. This high demand can be realised only in stages. The commercial products CHLORTHION® and METASYSTOX®[1]) are milestones along the road towards achieving the above aim. Due to its low order of mammalian and fish toxicity (see values below), CHLORTHION is especially suitable for controlling mosquitoes and for cover treatments.

Mammalian toxicity LD_{50}	=	625 mg/kg rat
Fish toxicity	=	5 ppm lethal after 18 hours
Fish toxicity	=	10 ppm lethal after 2 hours

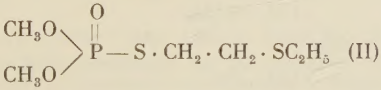
METASYSTOX, a monothiophosphoric ester, proved to be an excellent product for combatting sucking insects, and which does not harm non-sucking beneficial insects.

The chemistry[2]) of the systemic substances of the "Systox" class is so novel and interesting that it invited further intensive research in this highly promising field. According to its chemical structure, METASYSTOX is a mixture of the isomers of dimethylthiophosphoric ester (I) and dimethylthiolphosphoric ester (II):



(I)

0,0-dimethyl-thiono-phosphoric-0-β-ethylmercapto-ethyl ester



(II)

0,0-dimethyl-thiol-phosphoric-S-β-ethylmercapto-ethyl ester

[1]) A. Henglein, G. Schrader and R. Mühlmann: Zt. f. Analyt. Chemie, 141 Vol. (1954) No. 4, 276—281;
A. Henglein and G. Schrader: Zt. f. Naturforschung 10 b (1955) No. 1, 12—19.

® = Registered trade mark of Farbenfabriken Bayer Aktiengesellschaft

[2]) G. Schrader: Angew. Chem. 69, No. 3 (1957), 86—90

A component common to the esters I and II is the β-ethylmercaptoethyl remainder:

$$-CH_2 \cdot CH_2 \cdot SC_2H_5$$

Substitution of the ethylmercapto group in ester II by other mercaptan remainders[3]) did not bring about any particular advantages.

The following thioethers were tested for their suitability:

$$-CH_2 \cdot CH_2 \cdot SCH_3$$

$$-CH_2 \cdot CH_2 \cdot SC_3H_7n$$

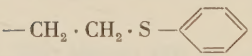
$$-CH_2 \cdot CH_2 \cdot SC_3H_7i$$

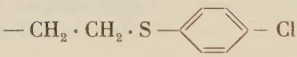
$$-CH_2 \cdot CH_2 \cdot SC_4H_9n$$

$$-CH_2 \cdot CH_2 \cdot SC_4H_9i$$

$$-CH_2 \cdot CH_2 \cdot SC_4H_9 \text{ sec.}$$

$$-CH_2 \cdot CH_2 \cdot SC_4H_9 \text{ tert.}$$





Shortening[4]), extending[5]) and branching[6]) of the alkylene remainder in the thioether group did not produce any positive improvement, either:

$$-CH_2 \cdot SC_2H_5$$

$$-CH_2 \cdot CH_2 \cdot CH_2 \cdot SC_2H_5$$

$$-CH_2 \cdot CH_2 \cdot CH_2 \cdot CH_2 \cdot SC_2H_5$$

$$-CH_2 \cdot \underset{\displaystyle CH_3}{\underset{|}{CH}} \cdot SC_2H_5$$

Introduction of the acetonitrile group[7]) in the thioether remainder also failed to improve the insecticidal properties:

$$-CH \begin{cases} CH_2 \cdot SC_2H_5 \\ CH_2 \cdot CN \end{cases}$$

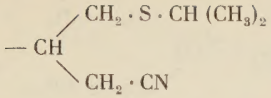
3) G. Schrader: DBP. 961.083 dated 3. 8. 1954.

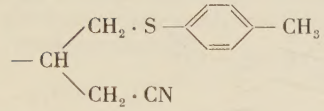
4) G. Schrader: DBP. 830.509 dated 24. 5. 1950.

5) G. Schrader: DBP. 836.349 dated 10. 5. 1950.

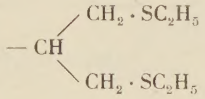
6) W. Lorenz and G. Schrader: DAS 1.035.394 dated 24. 11. 1955.
W. Lorenz and G. Schrader: DAS 1.035.958 dated 26. 11 1955.

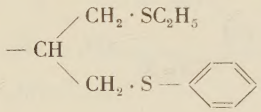
7) A. Dörken and G. Schrader: DAS 1.036.846 dated 21. 12. 1956.

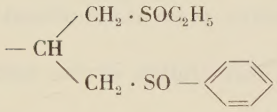


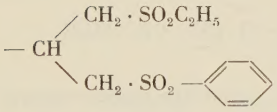


The introduction of a second alkyl- or arylmercapto remainder[8]) was likewise unsuccessful:









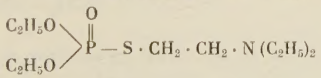
An attempt was also made to introduce the "Inferno" remainder[9])

$$-CH_2 \cdot CH_2 \cdot N(C_2H_5)_2$$

in the thioether group of METASYSTOX [10]). The resultant esters then have the following composition:

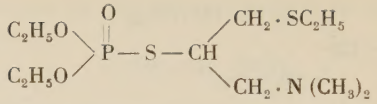
8) A. Dörken and G. Schrader: DAS 1·063·153 dated 10. 12. 1956.

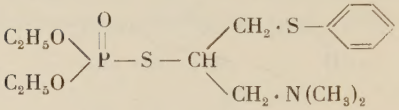
9) A phosphoric ester has been developed by the Imperial Chemical Industries known under the names "Inferno", "Amiton" and "Tetram", which, instead of an alkylmercapto remainder, contains the dialkylaminoethyl remainder:

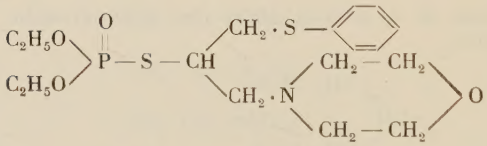


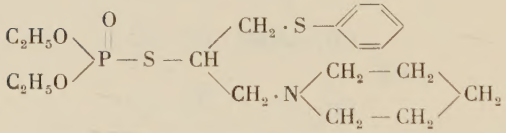
cf. E. P. 738·839 dated 19. 11. 1952;
R. Ghosh and J. F. Newman: Chem. and Ind. (1955), 118

10) G. Schrader and A. Dörken: DAS 1·027·208 dated 19. 5. 1956



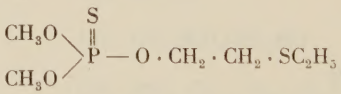






The prepared compounds have a very good insecticidal action but their high order of mammalian toxicity prevents practical use of this class of substances.

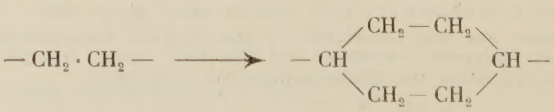
Let us return again to the constitution of the active ingredient META-SYSTOX given under I:



All attempts to bring about a definite improvement by substitution of the ethylene group

$$— CH_2 \cdot CH_2 —$$

had failed. We, therefore, abandoned the possibility of further substitutions in the ethylene group but we did give consideration to the question as to what effect might be produced by extending the linear ethylene group to form a ring-shaped structure:



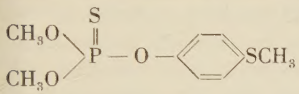
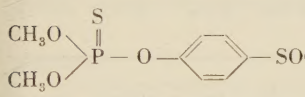
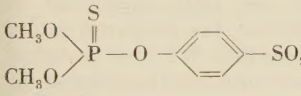
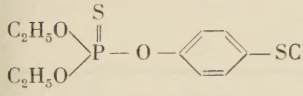
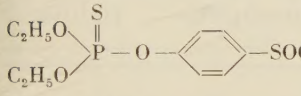
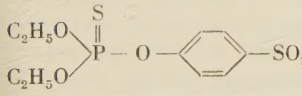
To support our theory, we first of all prepared 4-methylmercapto-1-oxy-benzene, described in the pertinent literature[11]), and reacted it with dimethyl thionophosphoric chloride:

11) E. Miller and R. R. Read: J. Am. Chem. Soc. 55 (1933), 1225.

(III)

Compound III became known under the internal number S 784. Its good insecticidal properties gave cause to continue research in this direction. The esters listed in Table 1 were prepared[12]):

Table 1:

Constitution	Toxicity[13]) to rat per os LD_{50}	Refractive index n_D^{20}	Insecticidal properties[14]) *Doralis fabae*		Fly larvae	
	10 mg/kg	1·5710	0·001%	90%	0·001%	100%
	10 mg/kg	1·5648	0·001%	40%	0·001%	100%
	10 mg/kg	1·5481	0·001%	85%	0·01%	100%
	5 mg/kg	1·5509	0·01%	60%	0·01%	100%
	10 mg/kg	1·5431	0·01%	20%	0·0001%	100%
	10 mg/kg	1·5542	0·1%	100%	—	—

The thionophosphoric esters listed in Table 1 are very interesting from the aspect of their insecticidal action but their high order of mammalian toxicity prevents them being used in crop protection.

12) G. Schrader and E. Schegk: Belg. Patent 556·009 dated 31. 3. 1956.

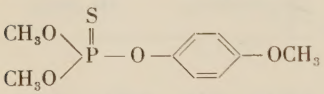
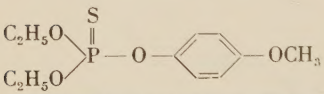
13) The toxic values were determined by Prof. Hecht at the Toxicological Laboratory of the Elberfeld Works.

14) The insecticidal properties were established by Prof. Unterstenhöfer and by Dr. Kükenthal and Dr. Jung.

In order to reduce the toxicity, we substituted the methylmercapto group by the methoxy group:

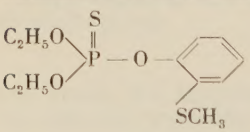
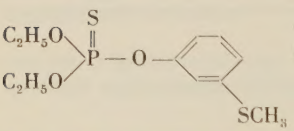
Table 2 gives the corresponding thionophosphoric esters that were obtained, together with details of their toxic and insecticidal properties:

Table 2:

Constitution	Toxicity to rat per os LD_{50}	b. p.	Insecticidal properties			
			Doralis fabae		Fly larvae	
	1000 mg/kg no symptoms	0·01/82°	0·1%	0 %	0·1%	0 %
	1000 mg/kg no symptoms	0·03/89°	0·1%	0 %	0·1%	60 %

It is evident from the values in Table 2 that by using the methoxy group instead of the p-positioned methylmercapto group, we fully succeeded in lowering the mammalian toxicity. Unfortunately, however, the prepared esters are of no value with respect to their insecticidal properties. We now reverted to methylmercaptophenol, and prepared the ortho- und meta-compounds. Table 3 provides information on the thiophosphoric esters we obtained:

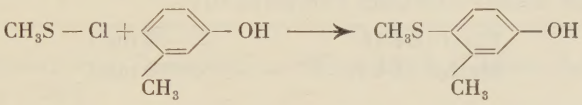
Table 3:

Constitution	Toxicity to rat per os LD_{50}	b. p.	Refractive index n_D^{20}	Insecticidal properties			
				Doralis fabae		Fly larvae	
	50 mg/kg	0·01/102°	1·5550	0·1%	70 %	0·001%	50 %
	50 mg/kg	0·01/102°	1·5528	0·01%	70 %	0·001%	50 %

It is perfectly obvious from the values in Table 3 that the ortho- or meta-positioned methylmercapto group offers no advantages. We now fell back again on the promising p-methylmercaptophenol and attempted, by means of suitable

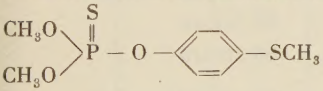
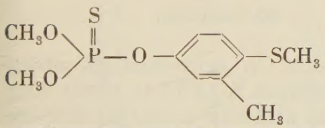
substitution, to bring the toxic values of the corresponding thionophosphoric esters within tolerable limits. Meanwhile, K. Wedemeyer and D. Delfs[15]) had developed a technical method for introducing the methylmercapto group in the p-position of phenols:

By means of this method, a number of substituted p-methylmercaptophenols became accessible. Inter alia, the 3-methyl-4-methylmercaptophenol was also prepared:



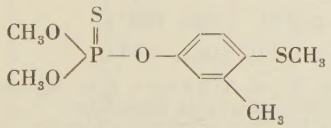
We phosphorylated this compound, and found, to our amazement, that by introducing the methyl group in the p-methylmercaptophenol, the toxicity of the corresponding phosphorylated compound was greatly reduced. Table 4 gives a comparison of the phosphoric esters of p-methylmercaptophenol and p-methylmercapto-3-methylphenol:

Table 4:

Constitution	Toxicity to rat per os LD_{50}	b. p.	Insecticidal properties *Doralis fabae*		Fly larvae	
	10 mg/kg	0·01/101°	0·001%	90%	0·001%	100%
	250 mg/kg	0·01/105°	0·001%	70%	0·001%	100%

Merely a glance at Table 4 shows that the toxicity has been reduced most considerably by the introduction of the methyl group, whereas the insecticidal properties have not been affected!

Thus, we had provisionally reached our goal. The product, known under the number S 1752[16]), and which has the following composition:



S 1752

15) Intermediates Dept. of Farbenfabriken Bayer Aktiengesellschaft, Leverkusen Works, DAS 1·063·177 dated 17. 4. 1957 and DAS 1·051·863 dated 13. 4. 1957.

16) cf. H. F. Jung: A New Phosphoric Ester Residual Insecticide with a Low Order of Toxicity, Bull. Org. mond. Santé 21 (1959), 215—221.

was now subjected to large-scale chemical, physical, pharmacological, toxicological and insecticidal tests.

1. Solubility and stability of S 1752 in water

S 1752, in a pure state, is a colourless liquid, slightly soluble in water. At room temperature, its solubility[17]) in water amounts to 54—56 mg/l. For the purpose of comparison, the corresponding figures for Ethyl-E 605[18]) and Methyl-E 605 are given below:

Solubility in water at room temperature

Ethyl-E 605	20—25 mg/l
Methyl-E 605	55—60 mg/l

To determine the rate of saponification, S 1752 was dissolved in equal parts of isopropyl alcohol and water, and heated to approximately 100 °C. Under these conditions, 50 % S 1752 is saponified in acid medium (pH 5—1·8) after 36 hours. In order to establish the alkaline saponification rate, 1/100 mol S 1752 was dissolved in 500 ml of isopropyl alcohol, 250 ml of water and 250 ml of 0,1 n caustic soda solution. When this mixture (pH 11) is brought to the boil, 50 % S 1752 is saponified in one hour and 35 minutes. To make this clearer, the corresponding values for E 605® are given below:

Rate of hydrolysis[19]) for Ethyl-E 605 and S 1752

		50 % saponification
Ethyl-E 605	acid medium at 100 °C	60 hours
S 1752	acid medium at 100 °C	36 hours
Ethyl-E 605	alkaline medium at 100 °C	25 minutes
S 1752	alkaline medium at 100 °C	95 minutes

These values indicate that S 1752 is saponified in acid medium about twice as quickly as E 605, whereas in alkaline medium S 1752 is about four times more stable than E 605. This high alkaline stability accounts for the typical residual effect[20]) featured by S 1752.

2. Boiling points, vapour pressure and volatility of S 1752

The active ingredient S 1752 can be distilled in high vacuum without decomposing. The following boiling points are obtained from the boiling-point curves plotted for S 1752:

b. p. 0·01 mm 105 °C
b. p. 0·0012 mm 60 °C

17) The solubility was determined by Dr. A. Dörken with P_{32}-labelled S 1752 at the Isotope Laboratory of Farbenfabriken Bayer at Elberfeld.

18) Determined by Dr. R. Mühlmann.

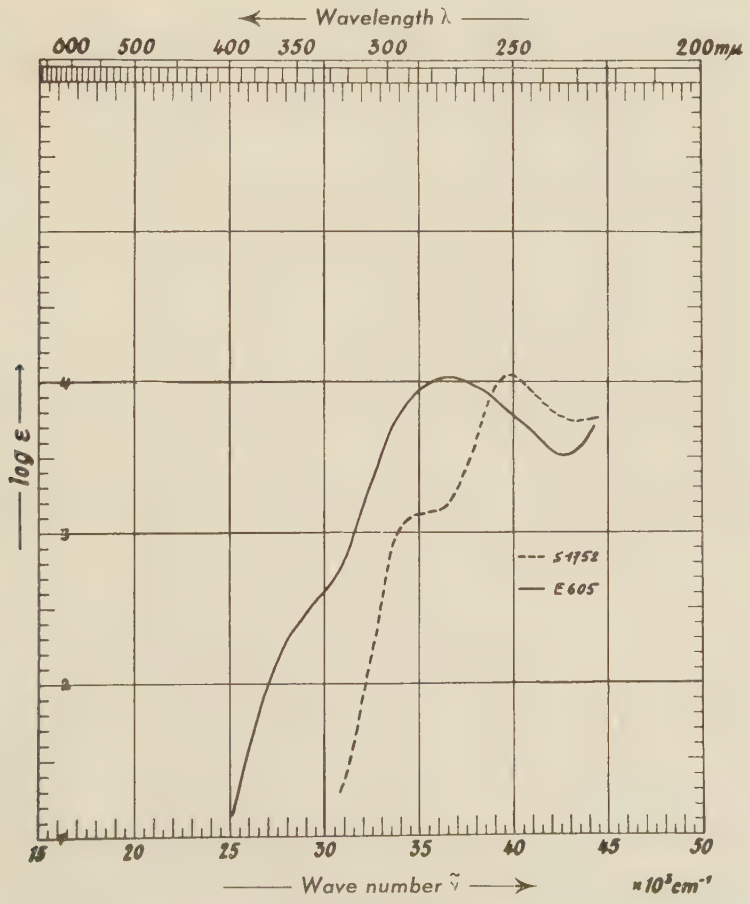
19) The values were determined at the Organic-Analytical Laboratory of Farbenfabriken Bayer Aktiengesellschaft, Leverkusen.

20) cf. H. F. Jung, loc. cit.

The vapour pressure and volatility at 20, 30 and 40 °C were calculated from the boiling-point curve for S 1752. The corresponding values for E 605 are also tabulated below for comparison:

	Vapour pressure		Volatility	
	S 1752	E 605	S 1752	E 605
20 °C	3×10^{-5}	$0{\cdot}57 \times 10^{-5}$ mm Hg	0·46	0·09 mg/cu. m.
30 °C	8×10^{-5}	$2{\cdot}27 \times 10^{-5}$ mm Hg	1·18	0·35 mg/cu. m.
40 °C	2×10^{-4}	$7{\cdot}04 \times 10^{-5}$ mm Hg	2·85	1·05 mg/cu. m.

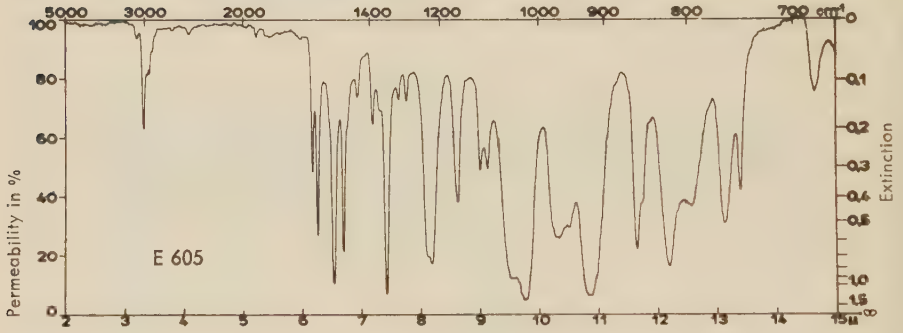
It is evident from these values that S 1752 is about three times more volatile than E 605.



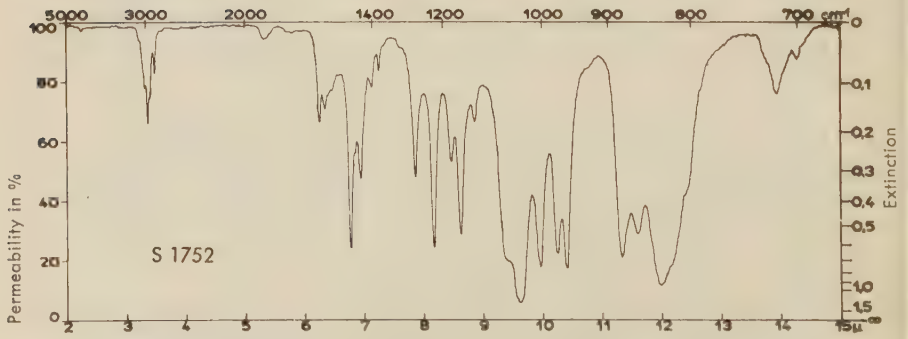
Solvent: Methanol

3. Ultraviolet and infra-red spectrum of S 1752

Whereas E 605 is broken down relatively quickly by short-wave light (ultraviolet rays), S 1752 is largely resistant to the whole spectrum of sunlight. The ultraviolet and infra-red spectra[21]) of S 1752 and E 605 are given below:



Dissolved 5% in CCl_4 and CS_2 Layer thickness 0·105 mm



Dissolved 5% in CCl_4 and CS_2 Layer thickness 0·105 mm

[21]) Plotted by Dr. W. Geiger, Physics Laboratory of Farbenfabriken Bayer Aktiengesellschaft, Elberfeld Works.

4. Toxicity[22]) of S 1752

S 1752 has a mean toxicity of 250 mg/kilo to the rat per os. The cutaneous toxicity of S 1752 is very slight. A dosage of 1000 mg/kilo was applied to the rat's skin but no symptoms were observed even after 4 hours. S 1752 is only slightly toxic also to other animal species, as shown by the following data:

Cat per os	100 mg/kg, no symptoms
Rabbit per os	250 mg/kg, died
Dog per os	100 mg/kg, survived, slight symptoms.

In comparison with E 605, Methyl-E 605 and GUSATHION®, the toxicity of S 1752 is extremely favourable:

Mean peroral toxicity to male rat

Ethyl-E 605	6·2 mg/kilo
Methyl-E 605	12·5 mg/kilo
GUSATHION	15·0 mg/kilo
S 1752	250·0 mg/kilo

5. Conversion of S 1752 in the living plant

The metabolites formed from the systemic insecticide SYSTOX in living plants have been largely clarified in studies conducted by Metcalf et. al, U.S.A.[23]) and by our own research workers[24]).

The redox system of the living plant converts a thioether group firstly into the sulphoxide and then into the sulphone:

$$CH_3S-R \longrightarrow CH_3SO-R \longrightarrow CH_3SO_2-R$$

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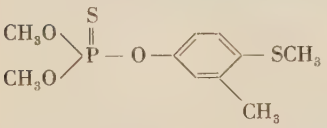
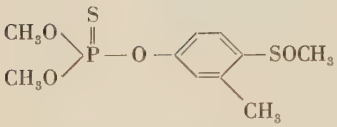
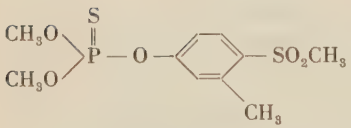
22) The values were determined by Prof. Hecht at the Toxicological Laboratory of Farbenfabriken Bayer Aktiengesellschaft, Elberfeld Works.

23) T. R. Fukuto, R. L. Metcalf, R. B. March and M. G. Maxon: Chemical Behaviour of Systox Isomers in Biological Systems, J. Econ. Ent. 48 (1955) 347—354; R. L. Metcalf, R. B. March, T. R. Fukuto and M. G. Maxon: The Behaviour of Systox-isomers in Bean and Citrus Plants, J. Econ. Ent. 47 (1954) 1045—1055; R. L. Metcalf, R. B. March, T. R. Fukuto and M. G. Maxon: The Nature and Significance of Systox Residues in Plant Materials, J. Econ. Ent. 48, (1955), 364—369.

24) R. Mühlmann and H. Tietz: Das chemische Verhalten von Methylisosystox in der lebenden Pflanze und das sich daraus ergebende Rückstandsproblem, Höfchen-Briefe 2 (1956), 116—140;
W. Wirth: Zur Wirkung System-insektizider Phosphorsäureester im Warmblüter-Stoffwechsel, Naunyn-Schmiedeberg's Arch. exp. Path. u. Pharmak. 234 (1958), 352—363.
G. Schrader: Die insektiziden Phosphorsäureester, Angew. Chem. 69 (1957), 86—90.

Since S 1752 contains a methyl-mercapto group, we prepared the corresponding sulphoxide and sulphone[25]). Table 5 provides data on the biological behaviour of the respective compounds:

Table 5:

Constitution	Rat per os LD_{50}	Effect on *Doralis fabae* Conc.	Mortality
 "S 1752"	250 mg/kg	0·001 %	70%
 "Sulphoxide"	125 mg/kg	0·001 %	80%
 "Sulphone"	125 mg/kg	0·1 %	85%

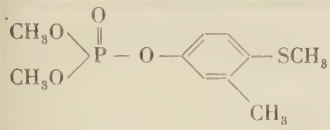
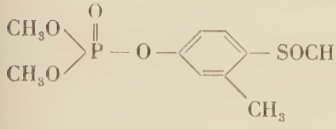
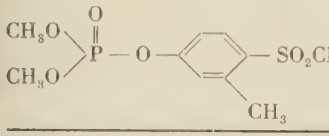
It is evident from the tabulated values that S 1752 becomes more toxic on changing over to the sulphoxide and sulphone stage. Whereas the sulphoxide is about biologically equal to the thioether, the insecticidal effect of the sulphone considerably decreases.

In order to register all conceivable conversion products of S 1752, the phosphoric esters of S 1752 given in Table 6 were prepared[26]):

25) E. Schegk and G. Schrader, Patent application Belgian Patent 579·006 of May 28, 1958.

26) cf. Advances in Pest Control Research, Vol. 1, P. 153, Interscience Publishers, Inc., New York 1957, published by R. L. Metcalf.

Table 6:

Constitution	Rat per os LD_{50}	Effect on *Tetranychus telarius* Conc.	Mortality
	100 mg/kg	0·01 %	98%
	50 mg/kg	0·01 %	95%
	25 mg/kg	0·01 %	95%

The values show that the oxygen analogue of S 1752 also becomes more toxic to mammals on being converted to the sulphoxide and sulphone form. The insecticidal effect is not reduced by the conversion to a higher stage of oxidation.

Summary

The development of the product S 1752 has provisionally brought to a close a series of tests which started with the systemic insecticide METASYSTOX.

The new product's low order of mammalian toxicity and its high resistance to alkalies make S 1752 extremely suitable for use in the public health and hygiene sector.

S 1752 has a marked systemic effect on large animals[27]), a feature which allows the conclusion to be drawn that this product promises to be most suitable also for controlling warble fly larvae and ticks in the veterinary sector.

BAYTEX® (S 1752, BAYER 29 493), a new hygiene insecticide from the group of low-toxic phosphoric esters

by H. F. Jung, H. Kükenthal and G. Technau

Almost half the world population lives in regions exposed to the hazards of malaria. Up to 1951, 250 million people became infected with malaria every year, and of this number 2 million died of the disease.

After the second world war, from 1946 to 1951, large-scale applications of the residual contact insecticide dichlorodiphenyltrichloroethane were carried out

[27]) Established by Dr. W. Behrenz of the Veterinary Dept. of Farbenfabriken Bayer Aktiengesellschaft, Elberfeld Works.

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in Greece, and proved fully successful in eliminating the *Anopheles* mosquito, the connecting link in the infection cycle of malaria.

The World Health Organization (WHO) thereupon decided, in cooperation with international and national public health services, to organize a campaign aimed at eradicating malaria all over the world, by applying residual contact insecticides. Thus, in 1951, the world-wide Malaria Eradication Plan was put into operation (WHO 1958a).

Paul Müller, the discoverer of dichlorodiphenyltrichloroethane, was awarded the Nobel Prize of Medicine in recognition of his services.

As a result of the wide use of dichlorodiphenyltrichloroethane and later of "Dieldrin", 116,165,000 people were again living in former malaria regions by 1958 — regions in which the danger of being infected by malaria had been completely eliminated.

This enormous success was brought about mainly by the fact that the mentioned active substances, in addition to being highly potent insecticides, also possessed the property of exeptional stability on porous and alkaline materials.

Unfortunately, it was found that out of the large number of insecticides developed in the following period, hardly any were equal to the mentioned chlorinated hydrocarbons with respect to residual efficacy on the substrates encountered in practical conditions. This was all the more regrettable in view of the fact that various pest strains proved to be capable of developing a considerable degree of resistance to these compounds, casting doubt on the success of the Malaria Eradication Plan in many areas. Due to the viability of the plasmodia in the human body, eradication of malaria is only possible provided reliable control of the disease-transmitting *Anopheles* mosquito is guaranteed over a period of 6 to 8 years.

Out of the 397,398,000 people who profited from the Malaria Eradication Plan, 25,000,000 lived at the end of 1958 in regions where the success of the scheme was seriously threatened by the development of resistant strains (WHO 1958b).

In order to overcome this resistance to chlorinated hydrocarbons, the obvious step is to use insecticides from other chemical groups, and which also have a different mode of action against the insect. Suitable for this purpose is the group of low-toxic, phosphoric esters although only to a restricted degree, for a number of cases have been encountered in which different active materials did not come up to expectation as residual insecticides due to their chemical and physical properties.

During the course of systematic research on phosphoric esters of low toxicity, conducted with the object of discovering suitable residual insecticides, a compound has been found which is already known to numerous experts as S 1752 (Bayer 29 493) or BAYTEX, and which appears to be the right product for closing the above-mentioned gap in the eradication of malaria, before it becomes too late.

BAYTEX (S 1752 or Bayer 29 493) was synthesized in the laboratory of Dr. G. Schrader (Schrader and Schegk, 1956); a report on its suitability

as a hygiene insecticide will be given in the following sections. The discovery of this compound, and its chemical, physical and toxicological data have already been dealt with in detail by Schrader in the preceding paper.

Insecticidal efficacy of BAYTEX in laboratory tests

Material and method

The insects used for the tests referred to in this section, were bred in the laboratory.

It is most essential for the reproducibility of biological tests, to have insects of the same origin, age and behaviour, otherwise the results may vary too greatly due to difference in vitality.

The best method proved to be to use certain larval stages of some insect species for testing purposes, rather than imagines, for the overall reaction of these larvae is more uniform than that of adults. Tables 1 to 4 therefore state which stages of the insect species involved were used for the tests.

All test insects used were kept in their breeding vessels until the start of the tests, and were at all times provided with adequate fodder. All blood-sucking species, with the exeption of *Culicidae*, were fed once with blood 1 to 2 days before the start of the tests. The *Culicidae* adults used for the tests were, on the other hand, only fed on glucose. The specimens used for the fish-toxicity tests were young fish of both sexes, measuring 2 to 2·5 cm in length. The mosquito larvae used were in the final instar, just prior to pupation.

Dishes, 9·8 cm in diameter and 6 cm deep, made of heat-resistant glass were used as test vessels; they were cleaned mechanically and chemically before each test. Afterwards, the equipment was heated up to 300 ^{0} C for three hours in a drier to remove any residues of active ingredient.

All other details concerning the implementation of the tests are given in the respective sections.

Effect on mosquito larvae and fish toxicity

Decisions as to whether and on what scale an insecticide can be used for controlling larvae of mosquitoes and other *Nematocera* species in waters, are frequently influenced also by the question as to what degree the respective insecticide has an adverse effect on fish in the waters that are to be treated or in adjoining waters.

Mass development of mosquito larvae admittedly takes places primarily in waters which, due to their smallness, lack of oxygen or short period of existence, usually offer no possibility for fish to breed and live in them, but in the tropics and sub-tropics *Poeciliidae* species of the genera *Gambusia, Mollienisia* and *Lebistes* do occur also in very small waters, and are undoubtedly the only effective natural enemies of mosquito larvae (King, 1944; Rockefeller Foundation, 1924). Simultaneous eradication of these fish during a control of mosquito larvae would soon reverse the success of such an operation.

Apart from these predators, consideration must also be given to algae-eating fish species bred chiefly in East Asia as food (e. g. *Chanos chanos* Forsk.); larvae of several *Chironomidae* species compete for fodder with these fish, and must therefore be controlled (Tang, 1959).

The suitability of an insecticide for controlling *Nematocera* larvae in fish waters is best assessed on the difference between the concentration of the insecticide that is just harmless to fish and the greatest dilution which, however, is still 100 % toxic to mosquito larvae. This difference is referred to as the safety factor, and may vary most considerably from one insecticide to the other.

In the following tests described here, BAYTEX active ingredient was dissolved in a small quantity of acetone, and diluted with tap water to the required concentrations. The solutions used for testing both the effect on mosquito larvae and fish toxicity were prepared in one operation in order to obtain equal conditions for genuine comparison.

For each test species, 2 glass dishes were each filled with 300 ml of solution of every concentration, and left open for 12 hours. At the end of this period, the test specimens were placed in the solutions, and mortality was registered after 24 and 48 hours. The following results are average values obtained for three replications of the above method. 10 fish and 40 mosquito larvae were used per concentration in each test; thus, for three replications, 30 fish and 120 mosquito larvae were used per test and concentration. In control tests using acetone less active ingredient, and tap water on its own, no mortality was recorded within the period of exposure and observation.

Fish toxicity:

Lebistes reticulatus Peters	10	ppm for 48 hrs. gives	100 %	mortality
	1	ppm for 48 hrs. gives	0 %	mortality

Effect on mosquito larvae:

Aedes aegypti L.	0·01	ppm for 24 hrs. gives	100 %	mortality
	0·005	ppm for 24 hrs. gives	70 %	mortality
	0·001	ppm for 24 hrs. gives	20 %	mortality
	0·0005	ppm for 24 hrs. gives	0 %	mortality
Culex pipiens L.	0·01	ppm for 24 hrs. gives	100 %	mortality
	0·005	ppm for 24 hrs. gives	30 %	mortality
	0·001	ppm for 24 hrs. gives	0 %	mortality

The tests showed that BAYTEX has a relatively low order of toxicity to fish, as compared with its high efficacy against mosquito larvae. A concentration of 1 ppm of BAYTEX active ingredient had no harmful effect whatsoever on young fish of the *Lebistes reticulatus* Peters species during exposure lasting 48 hours. The behaviour of the test individuals was perfectly normal also during the subsequent observation period of 14 days. On the other hand, a concentration of as low as 0·01 ppm proved lethal after less than 24 hours to *Aedes aegypti* and *Culex pipiens* larvae that were about to pupate. From these results, it can be concluded that BAYTEX must be applied at an overdosage 1000 times stronger than a concentration which gives 100 % mortality of mosquito larvae, for it to be harmful to fish. Therefore, when instructions on the application of BAYTEX formulations for controlling mosquito larvae (e. g. BAYTEX Granulate) are carefully observed, such applications will be absolutely harmless to fish.

Insecticidal potency

Tables 1 to 4 provide information on the insecticidal potency of BAYTEX (S 1752) against 12 *Orthoptera*, 10 *Coleoptera*, 10 *Diptera*, 3 *Rynchota* and one *Ixodidae* species. The following procedure was used in all these tests:

Filter papers were impregnated with acetone solutions of BAYTEX ingredient at the required concentrations. After the solvent had evaporated, the filter papers were placed in glass dishes, colonized with test insects, and covers placed over the dishes. The filter papers were either left dry or moistened

Table 1: Results of laboratory tests using BAYTEX (S 1752) against different *Orthoptera* species.

Test species	Dosage of active ingredient in mg per 1 m^2 filter paper	Period of exposure	Percentage mortality of test individuals at end of given exposure period
Blaberus gigantens L.	50	48 hrs.	100
(3rd stage larvae)	5	72 hrs.	0
Blaberus fuscus L.	50	48 hrs.	100
(3rd stage larvae)	5	72 hrs.	80
	0·5	72 hrs.	0
Blattella germanica L.	50	24 hrs.	100
(Adults)	5	72 hrs.	0
Blatta orientalis L.	50	48 hrs.	100
(4th stage larvae)	5	72 hrs.	0
Henschoutedenia flexivitta Walk.	50	48 hrs.	100
(3rd and 4th stage larvae)	5	72 hrs.	0
Jagrehnia testacea Br. W.	50	72 hrs.	100
(3rd and 4th stage larvae)	5	72 hrs.	0
Periplaneta americana L.	50	48 hrs.	100
(3rd and 4th stage larvae)	5	72 hrs.	0
Periplaneta australasiae L.	50	48 hrs.	100
(3rd and 4th stage larvae)	5	72 hrs.	0
Rhyparobia madeirae F.	50	24 hrs.	100
(3rd stage larvae)	5	72 hrs.	0
Gryllus domesticus L.	5	48 hrs.	100
(3rd and 4th stage larvae)	0·5	72 hrs.	50
	0·05	72 hrs.	0
Locusta migratoria L.	50	24 hrs.	100
(4th and 5th stage larvae)	5	48 hrs.	60
	0·5	72 hrs.	0
Schistocerca gregaria Forsk.	50	24 hrs.	100
(4th and 5th stage larvae)	5	72 hrs.	0

Table 2: Results of laboratory tests using BAYTEX (S 1752) against different *Coleoptera* species

Test species	Dosage of active ingredient in mg per 1 m^2 filter paper	Period of exposure	Percentage mortality of test individuals at end of given exposure period
Acanthoscelides obtectus Say.	5	24 hrs.	100
(Adults)	0·5	48 hrs.	0
Anthrenus scrophulariae L.	5	24 hrs.	100
(3rd and 4th stage larvae)	0·5	48 hrs.	60
	0·05	48 hrs.	0
Calandra granaria L.	50	24 hrs.	100
(Adults)	5	48 hrs.	60
	0·5	48 hrs.	0
Dermestes frischi Kugel	50	24 hrs.	100
(Adults)	5	48 hrs.	80
	0·5	48 hrs.	10
Dermestes lardarius L.	50	24 hrs.	100
(Adults)	5	48 hrs.	60
	0·5	48 hrs.	0
Oryzaephilus surinamensis L.	0·5	24 hrs.	100
(Adults)	0·05	48 hrs.	100
	0·005	48 hrs.	0
Tenebrio molitor L.	50	24 hrs.	100
(3rd and 4th stage larvae)	5	48 hrs.	70
	0·5	48 hrs.	10
Tribolium castaneum Herbst	0·5	24 hrs.	100
(Adults)	0·05	48 hrs.	70
	0·005	48 hrs.	0
Tribolium destructor Uyttenb.	5	24 hrs.	100
(Larvae)	0·5	48 hrs.	20
	0·05	48 hrs.	0
Trogoderma granarium Everts	5	24 hrs.	100
(3rd and 4th stage larvae)	0·5	48 hrs.	90
	0·05	48 hrs.	0

with tap water according to the requirements of the test individuals. The tests were evaluated after an exposure period of up to 8 hours for the *Diptera* species, and after 24, 48 and 72 hours for the other *Arthropoda* species. The length of each maximum period of exposure was fixed in relation to the natural vitality of the respective test species.

10 test individuals were used in each experiment, thus 30 individuals per species and concentration for three replications. Tests for which more than 10 % mortality was recorded in the controls (untreated and acetone less active ingredient), were not evaluated.

Table 3: Results of laboratory tests using BAYTEX (S 1752) against different *Diptera* species

Test species	Dosage of active ingredient in mg per 1 m^2 filter paper	Period of exposure	Percentage mortality of test individuals at end of given exposure period
Aedes aegypti L.	50	1 hr. 30 min.	100
(Adults)	5	2 hrs. 30 min.	100
	0·5	8 hrs.	100
	0·05	8 hrs.	0
Culex pipiens L.	50	1 hr. 20 min.	100
(Adults)	5	2 hrs. 10 min.	100
	0·5	8 hrs.	100
	0·05	8 hrs.	0
Calliphora erythrocephala Meig.	500	1 hr. 30 min.	100
(Adults)	50	8 hrs.	100
	5	8 hrs.	40
Chrysomyia chlorophyga Wied.	50	2 hrs. 50 min.	100
(Adults)	5	8 hrs.	100
	0·5		
Chrysomyia putoria Wied.	50	1 hr. 40 min.	100
(Adults)	5	8 hrs.	100
	0·5	8 hrs.	10
Drosophila melanogaster Meig.	0·5	2 hrs. 30 min.	100
(Adults)	0·05	4 hrs.	100
	0·005	8 hrs.	40
	0·0005	8 hrs.	20
Fannia canicularis L.	50	1 hr. 45 min.	100
(Adults)	5	8 hrs.	100
	0·5	8 hrs.	20
Lucilia sericata Meig.	50	3 hrs. 55 min.	100
(Adults)	5	6 hrs.	100
	0·5	8 hrs.	0
Musca domestica L.	50	1 hr. 45 min.	100
(Adults)	5	3 hrs.	100
	0·5	8 hrs.	100
	0·05	8 hrs.	20
Musca domestica L.	5	72 hrs.	100
(3rd stage larvae)	0·5	72 hrs.	50
	0·05	72 hrs.	10
Phormia regina Meig.	50	3 hrs. 30 min.	100
(Adults)	5	8 hrs.	90
	0·5	8 hrs.	0

The results of these tests revealed that dosages of BAYTEX active ingredient ranging between 50 and 0·5 mg per 1 sq. metre of filter paper reliably kill 36 species of *Arthropoda* within the given periods of exposure.

Table 4: Results of laboratory tests using BAYTEX (S 1752) against different *Rhynchota* species and one *Ornithodorus* species

Test species	Dosage of active ingredient in mg per 1 m^2 filter paper	Period of exposure	Percentage mortality of test individuals at end of given exposure period
Cimex lectularius L.	5	24 hrs.	100
(Adults)	0·5	24 hrs.	40
	0·05	24 hrs.	0
Rhodnius prolixus Stahl	50	48 hrs.	100
(3rd and 4th stage larvae)	5	48 hrs.	0
Triatoma infestans	50	48 hrs.	100
(3rd and 4th stage larvae)	5	48 hrs.	0
Ornithodorus moubata Murray	50	48 hrs.	100
(Adults)	5	48 hrs.	0

Residual efficacy on materials of different porosity and alkalinity

Wettable powders are known to be the best formulations suitable for producing insecticidally effective residual deposits; an important factor that must be given full consideration in order to obtain maximum efficacy from these formulations is their particle size (Hadaway and Barlow, 1951, 1952, 1952b; Treboux, 1957). The use of emulsion formulations is recommended only for application on less porous materials such as wood, paper, coats of oil paint etc., the reason being that emulsions penetrate into porous materials to a far greater extent than do wettable powders, and thus may lose their insecticidal effect.

For the following tests, we used a 40 % wettable powder formulation (4894a) with the following physical data:

Suspension property	Particle analysis
	$< 1{\cdot}2\ \mu$ = 34·1 %
	1·2 —10 μ = 34·6 %
> 75 % after 30 minutes	10 —20 μ = 11·4 %
	> 20 μ = 19·9 %

The experimental method was varied according to the test species used; this will be explained in more detail in the discussion of the different tables

Table 5: Tests of the residual effect of spray deposits of S 1752 (Bayer 29493) on different materials

Test insects: *Aedes aegypti* L. No. of test insects per test: 40

Preparation	Dosage of active ingredient applied per sq. m. of surface, in gr	Material treated	Percentage mortality of test insects after 24 hours following 1 hour of exposure to deposits					
			Age of deposit					
			1 week	2 weeks	1 month	2 months	3 months	4 months
S 1752 = (Bayer 29493) 40% Wettable Powder	1·7	Loam, lime-coated	100	100	100	100	90	40
	1·7	Loam, not lime-coated	100	100	100	100	70	50
	1·5	Burnt clay, lime-coated	100	100	100	100	95	60
	1·5	Burnt clay, not lime-coated	100	100	100	100	100	100
	1·2	Plywood, lime-coated	100	100	100	100	100	100
	1·2	Plywood, not lime-coated	100	100	100	100	100	100
	1·0	Glass, lime-coated	100	100	100	100	100	100
	1·0	Glass, not lime-coated	100	100	100	100	100	100
Control	untreated	Loam, lime-coated	0	10	0	0	0	0
		Loam, not lime-coated	10	0	0	0	0	0
		Burnt clay, lime-coated	0	5	0	10	0	0
		Burnt clay, not lime-coated	0	0	0	0	5	0
		Plywood, lime-coated	0	0	0	0	0	10
		Plywood, not lime-coated	0	0	0	10	0	0
		Glass, lime-coated	0	0	0	0	20	0
		Glass, not lime-coated	15	0	0	0	0	0

although the test procedure described hereinafter can be taken as the standard method.

Four types of material in the form of sheets with surface area of 200 square centimetres were used as substratum, these being loam, burned clay, plywood and glass. All these materials were included in the tests less a lime coating as well as in a lime-coated condition, i.e. before the insecticide was applied, some of them were coated with a 15 % suspension of slaked lime at a dosage of approx. 150 gr per square metre in order also to assess the resistance to alkalies of the residual deposits that were tested. The aqueous suspensions of the formulation were sprayed on to the materials used by means of an atomizer device. The quantities of active substance actually deposited on the materials were determined by weighing prior to and immediately after spraying.

Procedure for testing residual deposits: The test insects were anaesthetized with carbon dioxide (CO_2) and then placed on filter paper not treated with active substance; the filter paper was then laid on the treated materials, and covered with a light-alloy ring (10·7 cm inside diameter; 1·8 cm inside depth)

Table 6: Tests of the residual effect of spray deposits of S 1752 (Bayer 29493) on different mat

Preparation	Dosage of active ingredient applied per sq.m. of surface, in gr	Material treated	1 day k. d. 95 after given no. of mins. ='or hrs. =h	morta % a expo of 8
S 1752 =	1·7	Loam, lime-coated	55'	10
(Bayer 29493)	1·7	Loam, not lime-coated	55'	10
40 % Wettable	1·5	Burnt clay, lime-coated	55'	10
Powder	1·5	Burnt clay, not lime-coated	55'	10
	1·2	Plywood, lime-coated	30'	10
	1·2	Plywood, not lime-coated	40'	10
	1·0	Glass, lime-coated	50'	10
	1·0	Glass, not lime-coated	55'	10
Control	untreated	Loam, lime-coated	> 8h	
		Loam, not lime-coated	> 8h	
		Burnt clay, lime-coated	> 8h	
		Burnt clay, not lime-coated	> 8h	
		Plywood, lime-coated	> 8h	
		Plywood, not lime-coated	> 8h	
		Glass, lime-coated	> 8h	
		Glass, not lime-coated	> 8h	

fitted on the top side with wire meshing (width of mesh 1·25 mm, no. of meshes per square centimetre = 64). As soon as the test insects had come out of the narcosis and assumed their normal deportment, the filter paper was removed from under the light-alloy ring so that the insects now rested on the treated material. At the end of the required time of exposure, the test insects were removed in the same manner from the treated materials, and transferred to glass dishes that contained no active ingredient, for 24 hour observation. Afterwards the percentage mortality was calculated. Each test was carried out with four replications and an untreated check on the different materials.

An advantage of using the atomizer device (made of glass) and the light-alloy rings with exchangeable wire cover, is that these objects can be cleaned very easily after each test to remove any residues of active substance. In the tests under discussion, this was done by cleaning them mechanically and chemically with solvents, and then heating them to 300 ° C for several hours, a procedure which requires that these objects must be very resistant to temperature.

st insects: *Musca domestica* L. No. of test insects per test: 40

Residual effect: age of insecticidal deposits									
1 week		1 month		2 months		3 months		4 months	
95 ren f = ' =h	mortality in % after exposure of 8 hrs.	k. d. 95 after given no. of mins. = ' or hrs.=h	mortality in % after exposure of 8 hrs.	k. d. 95 after given no. of mins. = ' or hrs.=h	mortality in % after exposure of 8 hrs.	k. d. 95 after given no. of mins. = ' or hrs.=h	mortality in % after exposure of 8 hrs.	k. d. 95 after given no. of mins. = ' or hrs.=h	mortality in % after exposure of 8 hrs.
	100	120'	100	160'	100	360'	100	> 8h	70
	100	120'	100	150'	100	420'	100	> 8h	90
	100	100'	100	120'	100	360'	100	480'	100
	100	75'	100	80'	100	120'	100	480'	100
	100	50'	100	100'	100	60'	100	120'	100
	100	60'	100	100'	100	90'	100	120'	100
	100	75'	100	100'	100	90'	100	120'	100
	100	70'	100	80'	100	70'	100	120'	100
	0	> 8h	0	> 8h	5	> 8h	0	> 8h	10
	0	> 8h	10	> 8h	0	> 8h	0	> 8h	0
	0	> 8h	10	> 8h	0	> 8h	0	> 8h	0
	0	> 8h	0	> 8h	0	> 8h	0	> 8h	0
	0	> 8h	0	> 8h	0	> 8h	0	> 8h	0
	0	> 8h	0	> 8h	0	> 8h	0	> 8h	0
	0	> 8h	0	> 8h	10	> 8h	0	> 8h	0
	0	> 8h	0	> 8h	0	> 8h	0	> 8h	0

The tests were carried out on insects of the following species:

1. *Aedes aegypti* L.
2. *Musca domestica* L.
3. *Rhodnius prolixus* Stahl
4. *Cimex lectularius* L.

The *Aedes aegypti* and *Musca domestica* L. specimens used were 2—3 day old female adults which had been fed with dextroglucose solution; the *Aedes aegypti* individuals had not had a blood meal. On the other hand, the imagines of *Rhodnius prolixus* and *Cimex lectularius* were fed with blood 1—2 days before the test. No distinction between sexes was made for the latter species.

Discussion of Tables 5 to 8

Table 5

The procedure followed for carrying out these tests fully corresponds with the described standard method. Further details are given in the table. The tests showed that imagines of *Aedes aegypti*, exposed for one hour to four-month-old wettable powder deposits of S 1752 (Bayer 29 493), are knocked down to an extent of between 40 % and 100 % during an observation period of 24 hours. 100 % mortality of the test insects was obtained on loam over a period of two months as reckoned from the application of the deposits, and over a period of 2—4 months on burned clay. The residual effect of the preparation on wood and glass is persistent for a period of four months.

Table 6

The test procedure here deviates from the standard method insofar as the test insects (*Musca domestica* L.) were kept in constant contact with the deposits for up to eight hours. The first column gives the time that elapsed until knock-down was obtained for 95 % of the test insects. The second column gives the percentage mortality of the test insects after exposure for 8 hours on the deposits.

The test showed that imagines of *Musca domestica*, after being exposed for up to eight hours on residual deposits of S 1752, are killed over a period of three months, when the materials are very absorptive (loam). For less absorptive materials (clay, wood, glass), the effect of the insecticidal deposits is persistent for four months.

Table 7

Due to the high resistance of the insects (*Rhodnius prolixus* Stahl) used in this test series, the insects were exposed to the insecticidal deposits for relatively long periods (24 to 48 hours). The tests were evaluated immediately following the given periods.

The tests showed that residual deposits of S 1752 act on *Rhodnius prolixus* for 2 to 3 months on absorptive materials like loam and clay, and for four months on less absorptive materials like wood and glass.

Table 7: Tests of the residual effect of spray deposits of S 1752 (Bayer 29493) on different materials

Test insects: *Rhodnius prolixus* Stahl No. of test insects per test: 20

Preparation	Dosage of active ingredient applied per sq.m. of surface, in gr	Material treated	Age of deposits: 1 week		2 weeks		1 month		2 months		3 months		4 months	
			% mortality after an exposure period of: 24 hrs.	48 hrs.	24 hrs.	48 hrs.	24 hrs.	48 hrs.	24 hrs.	48 hrs.	24 hrs.	48 hrs.	24 hrs.	48 hrs.
S 1752 = (Bayer 29493) 40% Wettable Powder	1·7	Loam, lime-coated	100	100	100	100	60	100	30	100	20	80	0	0
	1·7	Loam, not lime-coated	100	100	100	100	40	100	20	100	20	60	0	0
	1·5	Burnt clay, lime-coated	100	100	100	100	60	100	70	100	40	70	0	20
	1·5	Burnt clay, not lime-coated	100	100	100	100	100	100	100	100	80	100	20	40
	1·2	Plywood, lime-coated	100	100	100	100	100	100	100	100	100	100	100	100
	1·2	Plywood, not lime-coated	100	100	100	100	100	100	100	100	100	100	100	100
	1·0	Glass, lime-coated	100	100	100	100	100	100	100	100	100	100	100	100
	1·0	Glass, not lime-coated	100	100	100	100	100	100	100	100	100	100	100	100
Control	untreated	Loam, lime-coated	0	0	0	0	0	0	0	0	0	0	0	0
		Loam, not lime-coated	0	0	0	0	0	0	0	0	0	0	0	0
		Burnt clay, lime-coated	0	0	0	0	0	0	10	10	0	0	0	0
		Burnt clay, not lime-coated	0	0	10	10	0	0	0	0	0	0	0	0
		Plywood, lime-coated	0	0	0	0	0	0	0	0	0	0	0	0
		Plywood, not lime-coated	0	0	0	0	0	0	0	15	0	0	0	10
		Glass, lime-coated	0	0	0	0	0	0	0	0	0	0	0	0
		Glass, not lime-coated	0	0	0	0	0	0	0	0	0	0	0	0

Table 8: Tests on the residual effect of spray deposits of S 1752 (Bayer 29493) on different materials

Test insects: *Cimex lectularius* L. No. of test insects per test: 40

Preparation	Dosage of active ingredient applied per sq. m. of surface in gr	Material treated	% mortality after exposure of 24 hrs. Age of deposits					
			1 week	2 weeks	1 month	2 months	3 months	4 months
S 1752 = (Bayer 29493) 40% Wettable Powder	1·7	Loam, lime-coated	100	100	100	100	100	20
	1·7	Loam, not lime-coated	100	100	100	100	100	20
	1·5	Burnt clay, lime-coated	100	100	100	100	100	100
	1·5	Burnt clay, not lime-coated	100	100	100	100	100	100
	1·2	Plywood, lime-coated	100	100	100	100	100	100
	1·2	Plywood, not lime-coated	100	100	100	100	100	100
	1·0	Glass, lime-coated	100	100	100	100	100	100
	1·0	Glass, not lime-coated	100	100	100	100	100	100
Control	untreated	Loam, lime-coated	0	0	0	0	0	20
		Loam, not lime-coated	0	0	0	0	0	0
		Burnt clay, lime-coated	0	20	0	0	10	0
		Burnt clay, not lime-coated	0	0	0	10	0	0
		Plywood, lime-coated	0	0	30	0	0	5
		Plywood, not lime-coated	0	0	0	5	0	0
		Glass, lime-coated	0	0	10	0	0	0
		Glass, not lime-coated	0	0	0	0	0	0

Table 8

In this test series, the test insects (*Cimex lectularius* L.) were exposed to the insecticidal deposits for 24 hours only, after which period the tests were evaluated.

The tests showed that insecticidal deposits of S 1752 applied to loam are residually effective against *Cimex lectularius* for three months. On clay, wood and glass, the residual effect persists for four months.

II. Insecticidal efficacy of BAYTEX in field tests

As indicated by the data in Tables 5 to 8, allowance was made in the laboratory tests for the influence exerted by the substrate on the active ingredient and formulation of an insecticidal wall deposit. It was also confirmed that the residual effect is largely influenced by the factors absorption and porosity (B e r t a g n a , 1959).

Fig. 1: Loam huts in the Belgian Congo, in their different stages of construction

In practice, the walls that have to be treated may be made of any of the following materials: Loam, clay (bricks), plaster, cement, concrete, stone, wood, bamboo mesh, liana mesh, banana leaves and finally straw or cane. Apart from the last five materials, the wall surfaces may be white-washed or not.

The house types illustrated in Figs. 1 to 8 provide an idea of the variety of materials available.

As these various materials are often used for building in one and the same colony, the selected dosage per square metre of surface area must be adapted to the most unfavourable material.

Fig. 2: Hut type — liana mesh covered with loam

Crystalline active ingredients like dichlorodiphenyltrichloroethane or MUSCATOX® have advantages over liquid active materials in this respect. BAYTEX is a liquid active ingredient. The formulation best suited for residual wall deposits, namely wettable powder, cannot be applied at concentrations over 40 % when the active ingredient is a liquid, and a very absorptive inert material must be used with it. In order to guarantee that the suspension property of the spray solution will be adequate, emulsifiers must be added.

Fig. 3: Treating a loam hut at Kinsuka, Belgian Congo

Depending upon the porosity of the material treated, the inert material will filter off on the surface retaining there the proportion of active ingredient held by it, whereas a certain quantity of the active ingredient contained in the water that penetrates into the wall will undoubtedly migrate in the wall; it is unlikely that there will be blooming on the surface, as in the case of crystalline active ingredients.

BAYTEX is a very stable active ingredient, thus accounting for the fact that in contrast to less stable phosphorus compounds, quantities of BAYTEX which microanalytically are relatively high and theoretically are still biologically effective, are to be found in the wall surface at a time when the biological effect normally disappears, whereas in the case of the less stable phosphorous compounds the proportion of active ingredient that has entered the wall undergoes a chemical breakdown there which is just as rapid as the disappearance of the biological effect on the surface.

Fig. 4: Loam huts near Thysville, Belgian Congo

40 % BAYTEX Wettable Powder, applied at a dosage of 1 gr active ingredient per square metre on loam, has proved to be residually effective for 26 weeks against *Culex fatigans,* a result which, considering the difficulties outlined above, is most impressive (Cerf/Lebrun, 1959b).

A. Effect against mosquitoes

The first field test of BAYTEX 40 was carried out at the end of 1957 at Léopoldville in the Belgian Congo. Here, *Culex fatigans,* in contrast to *Anopheles,* had especially eluded the very intensive and systematic treatment of the town area with chlorinated hydrocarbons carried out over a number of years, by rapidly developing resistant strains (Cerf/Lebrun, 1959). Furthermore, in Léopoldville there were thousands of native huts built of very unfavourable materials such as tamped loam or raw plaster. Apart from the selected dosage

Fig. 5: Bamboo mesh and white-washed loam, Huayto alto, Peru

Fig. 6: Wickerwork and loam, Chimbote, Peru

of only 1 gr of active ingredient per sq. metre, extremely stringent testing conditions prevailed with respect to the resistance of the test insects and the difficulties presented by the nature of the building materials.

Thanks to the kind assistance of the Wanson Institute in Léopoldville, the susceptibility of *Culex fatigans* to BAYTEX was first established in the labo-

Fig. 7: Coloured plaster, Léopoldville, Belgian Congo

Fig. 8: Crude brickwork, Léopoldville, Belgian Congo

ratory. It was found that the Léopoldville populations of *Culex fatigans*, despite maximum resistance to chlorinated hydrocarbons, were completely susceptible to BAYTEX. The BAYTEX values obtained in the tests on *Culex* larvae, 0·005 ppm for LD_{50} and 0·01 ppm for LD_{100}, were in the same order of magnitude as the values obtained in the Leverkusen laboratory tests on the fully susceptible *Aedes aegypti* strain.

Tables 9 and 10 provide data on the resistance of *Culex fatigans* in Léopoldville to chlorinated hydrocarbons and their susceptibility to a number of organophosphorus compounds and carbamates.

Table 9: LD_{50} of some insecticides to *Culex fatigans* of normal susceptibility, according to Cerf/Lebrun, 1959a

Author	Place	Insecticide (conc. in ppm) Dichlorodiphenyl-trichloroethane	HCH	Diedrin
Wharton	Malacca	0·145	0·014	0·003
Reid	Malacca	0·22	0·026	0·006
Elliott	Nigeria	0·03	0·05	0·005

Table 10: LD_{50} and LD_{90} of different insecticides to *Culex fatigans* larvae in Léopoldville, Belgian Congo, according to Cerf/Lebrun, 1959a

Insecticide	LD_{50} (ppm)	LD_{90} (ppm)
Dichlorodiphenyltrichloroethane	4·2	16
HCH	0·08	0·56
Dieldrin	0·15	0·46
Hercules 426	0·52	13 p.
MUSCATOX	0·016	0·05
DIPTEREX	0·069	0·23
Malathion	0·07	0·17
Diazinon	0·06	0·18
CHLORTHION®	0·007	0·019
Korlan	0·018	0·036
Mylone 85 W	2	—
Pyrolan	5	—
Bayer S 1752	0·004	0·01

After the susceptibility had been established, a small-scale primary field test was carried out. It was planned to conduct this test on a native hut that had not previously been treated with insecticides, and as such a hut could not be found in Léopoldville, the trial had to take place at the village of Kinsuka 25 kilometres away. The construction of the huts at this village and the way they were treated inside and outside, is illustrated in Figs. 1 and 2.

Kinsuka is located in the area covered by the hygiene control service of the Wanson Institute. At intervals of 7 days, insects present on the walls inside the test huts are trapped. This work is done in the morning, and use is made of the conventional type of suction tube. The trapped insects are

indentified and registered at the Wanson Institute, thus providing a regular check on the vector situation, with details of abundance and species. Table 11 gives the numbers of *Culex fatigans* trapped in the test hut during the period prior to the test.

Table 11: No. of *Culex fatigans* trapped at Kinsuka before start of test.

Date	No. of trapped *Culex fatigans*
31. 10. 1957	5
6. 11. 1957	8
13. 11. 1957	33
20. 11. 1957	6
27. 11. 1957	32
5. 12. 1957	34
12. 12. 1957	61
19. 12. 1957	34
26. 12. 1957	48
2. 1. 1958	23
9. 1. 1958	18
23. 1. 1958	21
30. 1. 1958	18
7. 2. 1958	19

The treatment was carried out on January 10, 1958 with a "Pompe Leman" type knapsack sprayer (Dobbins 8055 nozzle; distance of nozzle to wall 20 centimetres; output approx. 2 litres per minute resp. approx. 100 ccs spray solution per sq. m). 185 square metres of wall surface were treated with 19 litres of a spray solution containing 1 % active ingredient (5 % wettable powder 20 %) = 1 gr/sq. m. The 100 ccs of spray solution were rapidly absorbed by the porous loam wall, and there was no running.

The test was evaluated by continuing the weekly checks. A number of *Culex fatigans* were also kept in cages as a control. It was found that when the test includes an untreated control inside the treated hut, by hanging the cages on a part of the wall surface especially left unsprayed for this purpose or by suspending them freely in the room, the control insects are also killed in the first four days after the application, apparently due to the fact that, after spraying, an "aerosol" fills the entire room, and settles on the untreated parts. It was not until after the first four days had elapsed that the test insects in the freely suspended control cages were found to survive, whereas in the cages hung up on the wall surface left untreated a percentage of the insects were still killed; this mortality clearly differed from the results of controls carried out in completely untreated comparison huts.

The cage method used in the primary test was also employed in the main tests carried out afterwards by the Wanson Institute in 480 huts, so that in view of this a description of this method will no doubt be of interest.

Wire cages measuring 20 × 20 × 20 centimetres, open on one side and provided with a muslin sleeve attached to one of the other sides, are hung up, as shown in Fig. 9, with the open side facing the treated wall surface. In order to force the insect to rest more on the test surface than on the sides of the cage, at least in the first few hours, the outer sides of the cage are lightly sprayed — before the cage is hung up — with a 33 % alcoholic solution of toluic acid-NN-diethylamide, known as a mosquito repellent. An advantage of such large cages is that the test insects have adequate air space which cannot become saturated with gas. On the other hand, large cages must be left on the

Fig. 9: Test cage measuring 20×20×20 cm (5 sides are treated with repellent)

wall longer than small cages. Experience so far accumulated has shown, however, that even when small cages are used, the test insects are in contact with the wall deposit for only half the length of time that the cage is hung up on the wall.

As the primary test was carried out solely with the object of trying out the experimental method, it was not checked by the cage method until the effect had died out. The weekly counts revealed that the huts which were treated on January 10, remained practically free of *Culex fatigans* until the middle of June.

On June 5, 1958, the Wanson Institute treated 403 houses in the native quarters of Léopoldville, and a further 80 houses on October 30, 1958.

Cerf/Lebrun communicated the results of this large-scale test carried out according to the methods described above, in a provisional report (Cerf/Lebrun, 1959b).

The data in Tables 12 and 13 which include the results of the cage test and the morning counts, show that after 26 weeks a 100 % effect is still given on cement and a 70 % effect on loam as based on the evaluation of the cage tests.

The counts showed the residual effect to be persistent for 33 weeks.

Table 12: Results of cage tests on *Culex fatigans* in Léopoldville, according to Cerf/Lebrun (1959b)

	Age of deposits in weeks	% mortality corrected according to Abbot
Loam walls	12	100
	13	95
	17	86
	26	70
Cement walls, white-washed	14	100
	26	100
	33	85

Table 13: Average no. of *Culex fatigans* trapped (mornings) per house in 1958 and 1959, according to Cerf/Lebrun (1959b)

Week	Untreated control houses Sectors				Treated control houses Sectors	
	I	III	IV	V	VI	II
					Before treatment:	
1958/15	—	4	4	5	2	2·5
16	3·5	2	2	4	4	—
17	—	2·5	2	6	2·5	2·5
18	—	4	5	4·5	2·5	6
19	3	15	3	5	4	4·5
20	3	7	—	5	5	3
21	3	1	4	4	6	4
22	9	11	—	13	4	3
23	10	5·5	12	8	3	3·5
24	3	15	5	6	0	—

Table 13 contd.

Week	Untreated control houses				Treated control houses	
	Sectors				Sectors	
	I	III	IV	V	VI	II
					Treated BAYTEX 20 %	
25	6·5	6	2	—	—	7
26	5	6	9	3	0	7
27	6	0	2	4·5	—	—
28	9	7·5	4	2·5	0	8
29	1·5	1·5	3	2	—	12
30	2·5	—	2	1	0	1·5
31	—	6	0·3	2	—	3
32	2·5	0·6	0·3	0·2	0	1·5
33	4·5	1·5	0·5	3	—	1
34	2	0	1	—	0	3
35	1·5	2	0·4	4	—	7
36	2	1·5	0·25	0	0	2
37	5	0	0	3	—	3
38	1·5	1·5	1	3·5	0	9
39	11	2	3	3	—	4
40	10	0	0	2·5	0	3
41	2	1·5	0	2	—	1
42	5	1	0·5	4	0	1·5
43	0·9	1	2·5	—	—	—
44	2·5	0·7	4·0	—	0	
						Treated BAYTEX 40
45	7·5	—	1	—	—	1·5
46	4	1	2·5	1	0	0
47	1	1·5	1·5	2·5	0	0
48	4	3	1	2	—	0
49	1	7	1·5	2	0·4	0
50	1·5	4	—	0·7	0·7	0
51	1	—	4	2	0·25	0
52	1	2·5	—	3	0·5	0
1959/ 1	1	8	—	3 4	0	0
2	3	6	0·15	3 2·75	0	0
3	1·5	2·5	6	1·5 2·8	0·4	0

When the new WHO test method with the bioassay conical chambers (Figs. 10 and 11) — which was still in the testing stage — became known, the wall deposits in Léopoldville were already 32 weeks old. As the effect was still satisfactory, the treated huts were tested by this new method. For an exposure period in the cages of 1 hour, 30 % mortality was recorded within 1 to 1½ hours,

also in the 33rd week. Tests in the 35th week produced 30 % mortality on cement, and 20 % mortality on loam (Lebrun/Cerf, 1959).

The use of the WHO method has shown also elsewhere, that there is a difference between the perceptible residual effect in the treated houses of one zone, as registered also by morning counts, and the cage tests with artificially controlled period of exposure.

When the exposure period is prolonged until 100 % knock-down of the test insects is obtained -- naturally always in comparison with the untreated control — the result will be considerably nearer to that obtained in the count.

On the other hand, the exposure period must be adapted to the natural behaviour of mosquitoes in inhabited huts before and after the "blood meal" because it is with this that the question of the transmission of diseases is associated.

The WHO has, therefore, commissioned a group of entomologists to study this question. (Studies of indoor behaviour patterns of mosquitoes).

It should also be mentioned with respect to the field tests conducted in the Belgian Congo that the inhabitants of the treated huts were not troubled in any way during the same period (26 to 33 weeks) by species of the genera *Blattella, Cimex* and *Culex* (Cerf/Lebrun, 1959b).

Table 14 gives the results so far available of large-scale field tests conducted to establish the residual effect of BAYTEX 40 against mosquitoes.

According to these results and on the basis of experience meanwhile accumulated in Africa and South America on the practical application of BAYTEX, a dosage of 1 gr of active ingredient (2·5 gr of BAYTEX 40) per square metre can be expected to give a residual effect lasting 5 to 6 months also on porous walls and against mosquito strains resistant to chlorinated hydrocarbons.

B. Effect against flies

Whereas spray controls of disease-transmitting mosquitoes are generally carried out in large areas by official national or international public health organizations, the control of flies is primarily undertaken by private bodies, and in the main consists of stable spraying. Thus, on the question of the effect given against flies, we have, instead of the results of several large-scale tests, the results of a series of tests carried out in different countries, chiefly in individual cattle stables. Whereas the walls of these stables were treated in the same manner as described for the trial controls of mosquitoes, the evaluation methods vary, which is only natural in view of the large number of tests undertaken. It is, however, possible to classify them in the following three groups:

Method 1

Cage test; in the tests we carried out ourselves, however, use was made of a circular metal cage closed at the top by a detachable wire gauze (Figs. 10 and 11).

Table 14: Residual effect of BAYTEX 40 against mosquitoes. Results of field tests

Mosquito species	Place	Dosage of active ingredient gr: sq. m.	Material treated	Test method	Mortality	Age of wall deposits in weeks
Culex fatigans	Léopoldville (Belgian Congo)	1	Loam	Cage test Léopoldville 24 hrs. exposure	100%	12
					70%	26
		1	Cement white-washed	„	100%	26
					85%	33
Culex fatigans	Ocumare de la Costa (Venezuela)	2	Loam	Enquestas	100%	> 20 [1]
Anopheles albimanus	El Salvadore (Salvadore)	1	Loam and straw	Cage test Salvador 1 hr. exposure	75—91%	12
Culex pipiens	Kurtekotten, Leverkusen (Germany)	1·5	White-washed plaster	Count	100%	> 8 [1]
Aedes aegypti (laboratory breeds)	Kurtekotten, Leverkusen (Germany)	1·5	White-washed plaster	Cage test Leverkusen	100%	> 8 [1]

[1]) Evaluation was not continued up to time the effect disappeared.

The flies, usually taken from laboratory breeds, are narcotized with CO_2 before being put into the cage. They are placed in the ring before it is attached to the wall. A narrow strip of adhesive paper is applied to prevent the flies from falling out. At the end of the exposure period, a round glass plate with a hole in the centre is pressed against the cage hanging on the wall, and the

Fig. 10: Left: Bioassay conical chamber of WHO, with suction tube for insects. Right: Light metal ring with wire gauze (Leverkusen), and CO_2 cylinder for narcotizing insects

Fig. 11: Left: Bioassay conical chamber of WHO. Right: Metal ring (Leverkusen) in test position on wall

Table 15: Initial effect and residual effect of BAYTEX 40 against flies. Results of field tests in cattle stables

Fly species	Place	Dosage of active ingredient in gr/m²	Material treated	Test method	Initial effect after application	Mortality	Age of wall deposit in weeks
Musca domestica	Kurtekotten, Leverkusen (Germany)	1·5	Plaster white-washed	I (1h exposure)	—	80%	8
Musca domestica, *Fannia canicularis*	Schönrath, Leverkusen (Germany)	1·5	Plaster white-washed	II	—	94%	8
Musca domestica, *Fannia canicularis*	Mönchshof, Opladen (Germany)	2	Plaster white-washed	II	—	88%	7
Musca domestica	Mantova (Italy)	2	Stone white-washed Ceiling Wickerwork	III	50% 20' 100% 30'	100%	> 6
Musca domestica	Mantova (Italy)	2	Stone white-washed	III	50% 15' 100% 25'	100%	> 6
Musca domestica	Runkerhof, Völs (South Tyrol)	2	Plaster white-washed	III	100% 10'	98%	16
Musca domestica	Moorhof, Völs (South Tyrol)	2	Plaster white-washed	III	100% 10'	100%	16
Musca domestica	Wörndle, St. Jacob (South Tyrol)	2	Stone white-washed	III	100% 30'	88%	12

Musca domestica	St. Gilles (France)	1	Wood	III	50% 30'	100%	8
		2	Brick white-washed	III	50% 30'	100%	8
Musca domestica	St. Gilles (France)	2	Plaster white-washed	III	50% 30'	100%	8
Musca domestica	St. Gilles (France)	1	Oil paint	III	—	100%	> 5
Musca domestica	Bessan (France)	2	Concrete	III	50% 30'	50%	16
Musca domestica	Bessan, Herault (France)	2	Stone white-washed	III	50% 30'	75%	16
Musca domestica	Bramois, Wallis (Switzerland)	2	Brick white-washed	III	50% 20' 100% 30'	100%	6 [1])
Musca domestica	Ormône, Wallis (Switzerland)	2	Hollow brick white-washed	III	50% 20' 100% 30'	100%	6 [1])
Musca domestica	Kirkely, Ås (Norway)	2	Concrete white-washed	II	—	95%	16
Musca domestica, Stomoxys calcitrans	Ashem, Ås (Norway)	2	Concrete white-washed	II	—	99%	> 8
Musca domestica, Stomoxys calcitrans, Fannia canicularis	Kjölsrud (Holmstrand)	2	Wood white-washed	II	—	95%	> 8
Musca domestica, Stomoxys calcitrans, Fannia canicularis	Holstadt, Ås (Norway)	2	Roof: Concrete white-washed Walls: Wood white-washed	II	—	100%	12

[1]) Fly populations highly resistant to phosphoric esters

flies are narcotized by blowing in CO_2 through the hole in the plate. The evaluation is then carried out in the laboratory. An advantage of these cages is that they can be cleaned to remove any residues of active ingredient by heating them or by applying acetone; this cannot be done with the WHO bioassay conical chambers because these are made of plastic material which cannot be heated, and absorb phosphoric ester.

Method II

The fly population is counted at certain intervals before the treatment and during the test in the test rooms and in an untreated control room, at ten spots especially frequented by flies; these locations are marked. The decrease in fly infestation brought about by the treatment, is expressed in % in relation to untreated = 100.

Method III

1 to 2 square metres of wall surface, especially frequented by flies, is marked off before the treatment. The average fly population on this area of surface is determined up to the time of the treatment. On the same area of surface, a check is carried out in the same manner one day after the treatment and subsequently at regular intervals, at least once a week. At the same time, the local fly infestation must be checked in adjoining control rooms for the duration of the test.

Table 15 gives the results of field tests so far carried out to determine the residual effect of BAYTEX 40 against flies. The table also contains data on the initial effect recorded in these tests. The Roman figures refer to the category of the respective test methods.

It is found that the action of BAYTEX 40 on flies is not so strong as its effect against mosquitoes. In practical application, a dosage of 2 gr of active ingredient per square metre (5 gr BAYTEX 40 W. P.) may be expected to give a residual effect lasting 8 to 10 weeks.

It should be mentioned that most of the tabulated values were obtained with flies resistant to chlorinated hydrocarbons.

The tests carried out in Wallis (Switzerland) were conducted on a material which is resistant to both chlorinated hydrocarbons and another organophosphorus compound.

Summary

The chlorinated hydrocarbons dichlorodiphenyltrichloroethane and dieldrin are the two most suitable residual insecticides on the market to date, for controlling pests that attack man. Their chemical and physical stability on porous and alkaline materials accounts for the importance they attach in the Malaria Eradication Plan of the WHO.

Recently a compound has been developed also from the group of organic phosphoric esters, known under the name of BAYTEX (S 1752, Bayer 29 493); this product is outstanding for its residual effect, a feature that deserves maximum interest especially in view of the increasing resistance of numerous hygiene pests to chlorinated hydrocarbon insecticides.

BAYTEX (S 1752, Bayer 29 493) has a very strong insecticidal action, and also a low order of mammalian and fish toxicity. Tables are given containing the results of laboratory and field tests conducted to establish the compound's insecticidal effect against 40 Arthropoda species. In laboratory tests, residual deposits of BAYTEX applied at dosages of 1 to 1·7 gr active ingredient per square metre on materials of varying porosity and alkalinity, were found to be persistent for several months against major pests that attack man.

In field tests, and as also confirmed by early practical experience, a dosage of 1 gr of BAYTEX per square metre, applied to porous and alkaline walls, gave a residual effect lasting several months against mosquitoes; a dosage of 2 gr of BAYTEX per square metre was persistent against flies for 8 to 10 weeks.

Test methods are described for determining the biological effect of hygiene insecticides in laboratory and field tests.

A close study is made of the question regarding the length of exposure periods in cage tests.

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LEBAYCID®, a new insecticide of low toxicity

by G. Unterstenhöfer

Biologically active phosphoric esters hitherto known follow, as Schrader (1) has shown, a uniform pattern of structure which generally serves as a working hypothesis for the discovery of new active materials. In view of this, it will be easy to understand the finding, largely substantiated by experiments, that the mechanism of action, in the sense of the primary point of application in the organism or, respectively, in the course of the toxic process, is to be regarded as most uniform for effective phosphoric esters. It is based on the inhibition of a group of esterases, in general, and the cholinesterase, in particular. Despite this uniform mode of action, the spectrum of activity and the toxicological characteristics of organic phosphorus compounds greatly vary, ranging from marked polytoxicity at the one extreme to sharply defined selectivity at the other. The general rule is that a wide spectrum of insecticidal activity is linked with a higher order of mammalian toxicity, and selectivity with low toxicity. This correlation applies only within a given dosage range for each active material whereby the dosages must be specified in absolute values for the characterization of mammalian toxicity, and for the spectrum of insecticidal activity in more or less narrow limits determined throughout by economic factors or by secondary effects (phytotoxicity). As a rule, the spectrum of activity also increases to the extent by which the dosages are raised.

It has, however, been established in large-scale systematic research on organophosphorus compounds that in this group of compounds members may by all means be present which, in addition to having a low order of mammalian toxicity, possess a wide spectrum of activity within a favourable dosage range. In conformity with the aforementioned rule, it was, however, discovered that these compounds, too, in contrast to the toxic, widely active substances, have a more or less marked optimum of action on a certain group of pests. From these findings, it was concluded that it ought to be possible to discover a substance of low toxicity yet possessing a wide spectrum of activity, which can be used as a versatile insecticide. It also had to be borne in mind, however,

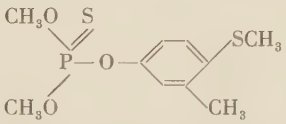
that the discovery of such an active ingredient, in face of the numerous biological and economic prerequisites that have to be satisfied, would have to be regarded as an accidental discovery in the statistical sense, in other words that it is a matter of a probability problem the solution of which becomes easier as the number of tested compounds increases. Following long series of tests, an active ingredient was found in LEBAYCID which combines the properties of an insecticide with a low order of toxicity and a wide spectrum of activity. In the meantime, an abundance of experimental data from laboratory, greenhouse and field tests has been accumulated, which justifies the presentation of a summarized survey of this active ingredient's properties and potencies. Further papers will be published later reporting on the effect produced by LEBAYCID for various uses in field tests.

This paper is based on the author's own tests, and is confined to the characterization of LEBAYCID as a crop protection product. Furthermore, the active ingredient has already proved its suitability as an insecticide for controlling insects that attack man (public health sector).

All dosages specified in the following are for pure active ingredient.

I. Chemical and physical properties

Chemically, LEBAYCID is 0,0-dimethyl-0-4-(methylmercapto)-3-methylphenylthiophosphate



The active ingredient is a brown liquid with an odour slightly reminiscent of garlic.

Molecular weight:	278·3	Volatility:	0·46 mg/m³ at 20 °C
Specific gravity:	1·250	Boiling point:	105 °C at 0·01 mm Hg
Refractive index:	n_D^{20} 1·5698		

Solubility: Soluble in most organic solvents; practically insoluble in water.

Stability: Exhaustive studies in the physics laboratory and in the organic-analytical laboratory of Farbenfabriken Bayer have revealed that for a phosphoric ester, the active ingredient possesses most remarkable stability in light, resistance to alkalies and hydrolysis-stability.

II. Mammalian toxicity

Although work on establishing the mammalian toxicity values of LEBAYCID is still in progress, it may already be said that this product can be classified among the active materials of low toxicity in the order of magnitude of dichlorodiphenyl trichloroethane, insofar as the acute toxicity is concerned. The LD_{50} to the rat per os is in the region of 230 to 250 mg/kg.

In rabbit ear tests, the active ingredient produced no symptoms of resorptive poisoning.

No symptoms of poisoning or irritation were observed in rabbits, rats and mice exposed for 2 hours to an atomized dosage of 1 cc/cu. m.

III. Insecticidal and acaricidal properties

1. Action on Diptera

The rule referred to earlier for insecticidal organophosphorus compounds whereby substances of low toxicity which have a wide spectrum of activity possess marked optima of use, also applies to LEBAYCID. We, therefore, exhaustively tested the active ingredient in the laboratory on fly species of economic importance, following the indication given in the initial experiments that a most striking property of the substance is its effect against Diptera. In keeping with customary practice, we first determined the limit concentrations by placing the flies and their larvae in closed cages on filter paper treated with solutions diluted in stages, and registering the percentages of mortality. The ovicidal effect on mangold fly (spinach leaf miner) was determined by spraying egg pods on beet leaves and registering the percentage of hatching. We used insect material we had bred ourselves for these tests, with the exception of olive fruit fly which was captured in the field. The test results are given in Table 1.

Table 1: Mean limit concentration of LEBAYCID effective against different species of *Diptera*.

Mangold fly *Pegomyia hyoscyami*	Mediterranean fruit fly *Ceratitis capitata*	Olive fruit fly *Dacus oleae*	*Drosophila melanogaster*	House fly *Musca domestica*
		Adults		
—	0·001 %	0·001 %	0·001 %	0·001 %
		Larvae		
0·0003 %	—	—	—	0·001 %
		Eggs		
0·002 %	—	—	—	—

These values show that LEBAYCID in a dilution of 1 : 100 000 still has a highly insecticidal effect against adults of important Diptera species. Larvae and eggs also react to the active ingredient to about the same degree, as far as could be established from the limited amount of test material. These exploratory tests showed that by virtue of the good effect given against adults, combined with the larvicidal and ovicidal potency, the active ingredient promises

to produce favourable results in further experiments to establish its efficacy under practical conditions.

With the technique employed in the first tests, the substance is allowed to act simultaneously as a stomach, contact and respiratory poison. The values thus recorded provided a good indication of the reaction of the different species and instars. However, if detailed information is required which may be very useful for comparative assessments of active materials, for the interpretation of field test results, for residue problems etc., it is the individual insect treatment which first provides a reliable basis for appraisal. Therefore, when active substances appear to be of interest, we usually follow up the determination of the limit concentration by carrying out topical individual insect treatments by means of the accepted test method, using a micrometer syringe, in order to determine exactly the mean lethal dosage per insect; for the purpose of extending our knowledge on the comparative effect on insects, on the one hand, and on mammals, on the other hand, we then convert these values to the quotients, mg poison/kilo body weight, generally used in toxicology. Our tests conducted from this aspect produced the results given in Table 2.

Table 2: Mean lethal contact poison dosage of LEBAYCID to different species of *Diptera* (LD_{50}).

Dacus oleae		*Ceratitis capitata*		*Pegomyia hyoscyami*		*Musca domestica*	
γ/insect LD_{50}	mg/kg LD_{50}	γ/insect LD_{50}	mg/kg LD_{50}	γ/insect LD_{50}	mg/kg LD_{50}	γ/insect LD_{50}	mg/kg LD_{50}
0·00006	0·012	0·0015	0·234	0·005	0·54	0·032	2·64

From these values, the following order is obtained for the percutaneous susceptibility of the different species:

Dacus oleae — *Ceratitis capitata* — *Pegomyia hyoscyami* — *Musca domestica*
1 ∼ 20 ∼ 40 ∼ 200

These values largely deviate for the different species from the data obtained in the determination of the limit concentration insofar as species-specific differences become manifest here. This means that the percutaneous resorption values for LEBAYCID apparently vary for the different fly species whereas the toxicity — provided all mechanisms of resorption are able to act — gives no indication of marked species-specific differences. These findings thus support the assumption occasionally expressed, which is based partly on observations and partly on considerations, that with the same method of applying active ingredient and the same sensitivity of the reactive system, varying dosages reach the site of action, whether due to introduced mechanical protective systems of varying strength or due to varying solubility relations, which in the unit of time lead to different concentrations at the site of action, or whether due to qualitative or quantitative, species-specific, fixed detoxication

processes. The results furthermore confirm that group-specific selectivity may be present also in active ingredients which like the organophosphorus compounds possess on principle a similar mechanism of action in poikilothermic animals and mammals. In these facts lies also the key to understanding that the organophosphorus compounds have provided highly efficient active ingredients for practically all purposes in the control of insects and mites in crop protection, in public health (hygiene) and in veterinary medicine, in the form of contact, stomach and respiratory poisons and systemic insecticides, which in their properties are able to adapt themselves to the respective technical requirements of pest control.

In order to clarify further the relation between stomach and contact poison action, we determined the peroral toxicity of LEBAYCID to *Ceratitis capitata* in an individual insect test by means of a method described in an earlier paper (2). The following values were obtained (Table 3).

Table 3: Mean lethal peroral dosage of LEBAYCID against *Ceratitis capitata*

Ceratitis capitata LD_{50}	
γ/insect	mg/kg
0·00116	0·23

This dosage largely corresponds to the percutaneous value obtained in the topical treatment so that especially for the Mediterranean fruit fly, peroral and percutaneous toxicity are in the same order of magnitude.

Further to the findings already given, it should be mentioned that as against the comparable organophosphorous compounds, LEBAYCID has a high rapidity of action which is independent of concentration to an unusual degree.

The potentially existing insecticidal effect of LEBAYCID against Diptera as described here was most convincingly borne out by the efficacy attained in the subsequent laboratory, greenhouse and field tests. Apart from the potency as such, the stability of the active ingredient — remarkable for a phosphoric ester — is also responsible for the attained effectiveness; this excellent stability is proved by a long residual effect which is especially important for the control of flies injurious to plants.

2. Action on Homoptera

Plant-sucking pests, especially aphids, attack practically all crops, and cause damage in the form of reduced yields and quality also when infestation is of a moderate degree. They occur with great regularity and thus require regular control. It is, therefore, necessary that insecticides also possess aphicidal potency, especially if they are to lay claim to versatile use. This all the more so as experience especially with chlorinated hydrocarbons — apart from a few exceptions — repeatedly shows that the lack of an effect against aphids necessitates carrying out special measures, in that not only does the population

density of aphids remain unaffected but also infestations frequently break out which are more severe that in the untreated controls. Precise indications of this are to be found every year in the Memphis Report published in the U. S. A. This report gives exact characteristics of the effect of the different insecticides for treatment of cotton.

In the biological tests of LEBAYCID, it was established that the active ingredient possesses considerable aphicidal potency. The following values were determined as the limit concentration (LD_{50}) for the two major aphis species *Myzodes persicae* and *Doralis fabae:*

Doralis fabae on *Vicia faba*	— 0·0007 %
Myzodes persicae on *Brassica spec.*	— 0·001 %

These values, therefore, are in the order of magnitude also determined for Diptera.

As expected, the residual effect was found to be dependent on concentration; the following values were obtained for 50 % mortality of *Myzodes persicae* in greenhouse tests, in which beets were artificially infested with the pest:

Concentration	Residual effect
0·025 %	2 days
0·05 %	5 days
0·1 %	10 days

These values were largely confirmed in later field tests against *Myzodes persicae* on potatoes.

Good results were obtained in laboratory and field tests against the relatively resistant apple aphis *(Doralis pomi).*

In the laboratory test of LEBAYCID on woolly aphis *(Eriosoma lanigerum),* the LD_{50} value was determined at 0·004 %.

LEBAYCID has only a moderate to weak effect against scale insects, as the following values reveal:

Pseudococcus citri	0·1 % / 80 %
Aspidiotus hederae	0·1 % / 60 %
Lecanium corni	0·03 % / 90 %
Lepidosaphes ulmi	0·03 % / 80 %

The relative resistance of the scale insects, however, is not brought about physiologically. It was established that by adding oil to LEBAYCID, the effect against scale insects was considerably improved. Apparently the power of LEBAYCID to penetrate through the scale integument is only moderate despite the good lipide-solubility of the active substance, and is improved by the action of oil.

3. Action on Lepidoptera

Diamondback moth (*Plutella maculipennis*): The first tests conducted with the object of an effect against caterpillars were carried out on diamondback moth, a species that can be bred continuously in the laboratory without any particular difficulty. This pest is moderately to strongly susceptible to the known products used for controlling caterpillars, and therefore is a very suitable test object for the technical development of a preparation. A value of 0·003 % was determined as the limit concentration (LD_{50}) for LEBAYCID.

Winter moth *(Cheimatobia brumata)*: In the test against winter moth caterpillars on potted apple trees, LEBAYCID gave a 40 % kill at a concentration of 0·002 %. On artificially infested trees sprayed with a concentration of 0·05 %, an efficacy of 60 % was recorded after 5 days. These results were confirmed in field tests.

Oak leaf roller moth *(Tortrix viridana)*: In the laboratory test LEBAYCID gave a 100 % kill of oak leaf roller moth caterpillars at the amazingly low concentration of 0·0004 %. The good effect of the product against this really susceptible species was confirmed in a small-scale field test.

Miscellaneous caterpillar species: After it had been established that LEBAYCID is most effective against moth caterpillars, small-scale field tests were carried out against different caterpillar species. In the following, a brief report is given of the results that were obtained.

Conclusive results were recorded at a concentration of 0·05 % against

Casebearer moth	*(Coleophora spec.)*
Ermine moth	*(Hyponomeuta spec.)*
Summer fruit tortricid	*(Capua reticulana)*
Pear leaf blister moth	*(Cemiostoma scitella)*

A most marked effect was also observed against codling moth *(Carpocapsa pomonella)* in the first exploratory tests. However, it will not be possible to give a final opinion on the actual effect that LEBAYCID can produce against this pest until large-scale field tests have been concluded.

It was most noticeable that the product only gave a moderate to weak effect against *Clysia ambiguella*. This result, however, should be treated with reservation because in the test a large number of the larvae had already entered the berries; thus the time of application here meant that extreme requirements had to be satisfied by the preparation.

4. Action on Coleoptera

According to our present level of knowledge the action of LEBAYCID against beetles is greatly species-specific. Just the same, appreciable effect is produced against several species, as the following values show:

Calandra granaria	0·004 %	—	100 %
Sitona spec.	0·01 %	—	100 %
Phytonomus variabilis	0·01 %	—	80 %
Apion pisi	0·01 %	—	80 %
Phyllobius piri	0·1 %	—	100 %
Rhyzotrogus solstitialis	0·001 %	—	100 %
Meligethes aeneus	dust	—	100 %

The effect registered against *Meligethes aeneus* was confirmed in field tests.

5. Action on Hymenoptera, Heteroptera and Thysanoptera

LEBAYCID applied at 0·001 % gave a 100 % kill of cherry sawfly *(Eriocampoides limacina)*.

Good results were obtained with dust formulations in laboratory tests against *Piesma quadrata*.

Banded greenhouse thrips *(Hercinothrips femoralis)* reacts well to LEBAYCID. A 100 % mortality was still recorded after 12 days in tests where beets were artificially infested with this pest and sprayed at 0·01 %.

6. Action on Mites

Organophosphorus compounds are all effective against mites, especially spider mites. In greenhouse tests conducted on a *Tetranychus telarius* strain of normal susceptibility, LEBAYCID produced a most appreciable acaricidal initial effect against postembryonic active stages at a limit concentration of 0·0008—0·002 %. An ovicidal effect was not registered.

Further tests on different species and strains then showed, however, that compared with the efficacy of marked miticides like METASYSTOX®, LEBAYCID should not be regarded as an acaricide from the aspect of control technique; rathermore the acaricidal action it does produce should be rated as a secondary effect. According to present-day standards, the effect against the fruit tree spider mite *(Paratetranychus pilosus)*, for example, must be rated as not being fully adequate, as field tests have revealed. The effect against *Tetranychus viennensis*, on the other hand, appears to be appreciably better. Further testing must be carried out before a final opinion is given on the acaricidal potency of LEBAYCID, especially its species-specific and strain-specific effect

Results of laboratory tests show that the product is ineffective against *Eriophyes ribis*. On the other hand, the broad mite *(Hemitarsonemus latus)* proved to be most susceptible; a 100 % kill was registered against it at a concentration of 0·001 %.

IV. Tolerance to plants

Although a final rating of the plant tolerance of an active ingredient which is to be widely used as an insecticide, cannot be given until it has already been applied on a large scale, especially in view of the existence of so many species and varieties and the frequent dependence of tolerance on the stage of plant growth and environmental conditions, phytotoxicity tests we have carried out to date with LEBAYCID on a number of plant species, in conjunction with our wide experience on the behaviour of organophosphorus compounds, do permit the conclusion to be drawn that the preparation is highly tolerable to plants. No injuries were observed whatsoever on the following plant species after treatment with 0·1 % LEBAYCID:

Apple (tested on 20 different varieties of economic importance), pear, cherry, peach, cotton, citrus, tomato, potato, beet, bean *(Phaseolus, Vicia)*, *Cineraria*, tulip, cyclamen, *Primula obconica*, *Philodendron erubescens*, *Rheocissus*, cabbage, lettuce, wheat, rye, melon, rose.

A certain sensitivity was observed on blooms of ornamentals such as tulips, *Primula obconica*, *Euphorbia pulcherrima* and carnations.

Summary

0,0-dimethyl-0-4-(methylmercapto)-3-methylphenylthiophosphate is the active component of the new insecticide LEBAYCID. A report is given in this paper on its biological efficacy.

1. With an LD_{50} to the rat of 230 to 250 mg/kg per os, LEBAYCID ranks among the organic phosphoric esters having a low order of mammalian toxicity.
2. The active material features a wide spectrum of activity as proved by the findings of tests on approximately 30 species of Diptera, Homoptera, Lepidoptera, Coleoptera and other orders of insects.
3. LEBAYCID possesses a moderate acaricidal potency.
4. LEBAYCID is highly tolerable to plants.

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Trial controls of olive fruit fly by means of LEBAYCID® in Crete

by W. Kolbe, Leverkusen

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The olive *(Olea europaea* L.) has been one of the most important cultivated trees of the Mediterranean zone for centuries past; olives and olive oil are today still important foods in all Mediterranean countries. The largest growing regions are in Spain, Italy, Greece and Portugal where olive oil and table olives are major export goods. Cultivation of olives has meanwhile spread from the Mediterranean to Australia, India, South Africa, North and South America. In 1956, world production of olive oil amounted to 1,220,000 tons, and the production figure for the same year in the Mediterranean countries was 900,000 tons (45).

The chief insect pest of olive is the olive fruit fly *(Dacus oleae* Gmelin). The damage caused by olive fruit fly — when no controls are carried out — ist estimated by Logothetis (20) at 600 million dollars for the Mediterranean zone; in Australia and America, this insect has not yet established itself as a major pest. Even when the different control measures, which will be discussed later, are carried out, the annual yield loss is nevertheless in the region of 25 %, according to the estimate of Logothetis (20).

In Greece where there are approximately 80,000,000* fruit-yielding olive trees with an average annual oil production of 120 000 tons, damage by olive fly is estimated by Kortzas (18) to be between 20 and 20 %, corresponding to an average value of 15 million dollars.

The olive fruit fly pierces the skin of the fruit with its ovipositor and lays its eggs in the flesh of the olive fruits. Each female lays between 200 and 250 eggs although usually only one egg per fruit. Consequently several eggs or larvae are present in one olive stem from different females. The larvae tunnel in the fruit flesh causing the fruit to disintegrate and fall. In summer, the larvae emerge after 2 to 3 days whereas in autumn at low temperatures they take longer to hatch out. Larval development lasts 14 to 30 days; the larvae do not migrate (from fruit to fruit), and thus pupate in the fruit. Pupal quiescence in the fruit is stated by Hennig (15) to last 10 days. The cycle of development for all stages is of varying duration, being dependent upon the temperature as is the number of broods or generations produced in a year. Generally, 4 to 6 generations occur (15, 20), and only 2 to 3 in regions with unfavourable climatic conditions (15, 20, 3); in some regions of North Africa, on the other hand, there is an uninterrupted sequence of broods (20). When the weather becomes cold in autumn, the full-grown larva leaves the fruit to pupate in the soil. The upper temperature limit is stated by Hennig (15) to be 32 to 34° C; maximum temperatures in the region of 40° C such as recorded in August 1958 in Crete, accompanied by a relative humidity of 30 %, bring about a complete collapse of the population. According to Hennig (15) the most severe damage is caused in summers with uniform relative humidity and mean temperatures of less than 24° C; according to Bredemann (10) this corresponds to the most favourable climate for the olive tree. This means that severe olive fruit fly infestation coincides with favourable local and cultivation conditions for olive trees, thus demonstrating the purposefulness of a control.

I. Survey of control methods

The olive fruit fly, already known to Theophrastus, has always been a scourge. According to Jegen (17) infested olives were prematurely harvested and pressed in former times in order to minimize damage to the crop, and in fact the author has observed that this method is still practised today in some parts. Jegen (17) goes on to report that former control operations were directed against overwintering pupae in the soil; ashes and lime were worked into the soil to eradicate the pupae, and woollen rags soaked in petroleum were buried under the soil surface; we know from present-day experience that such methods offered no prospect of great success. A heat-wave in summer, with temperatures ranging from 35 to 40° C, on the other hand, frequently led to a complete collapse of the *Dacus* population in the olive-growing regions; depending upon the degree of infestation already prevailing, this resulted in a reduction of crop damage to a greater or lesser extent (3, 4, 44).

Since the beginning of the present century, work has continued intensively in a effort to solve the *Dacus* problem, furthered by the provision of state funds, the establishment of institutes and by prize competitions introduced to give added incentive. Also since the turn of the century, the practice has been

* According to Triantaphyllidis (40 a), there were 73,565,050 olive trees in Greece in 1959.

adopted of controlling the olive fruit fly with liquid arsenic baits; this method originated in Italy. The arsenic baits consisting of molasses and sodium arsenate, which were initially developed for olive fruit fly control by de Cillis and later by Berlese, are sprayed onto the boughs growing on the south side of the trees. In the application of arsenic baits, exploitation is made of the fact that the flies leave the puparium with immature ovaries. According to Berlese, the olive fruit fly begins to lay its eggs 8 to 10 days after emerging, and during this period the flies seek sugary liquids for which they show a marked partiality as food because it is of utmost importance to the development of the imagines (39). It must, therefore, be the aim of liquid arsenic bait applications — known today in olive-growing as the Berlese method — to kill

Fig. 1: The olive fruit fly pierces the skin of the fruit with its ovipositor and lays its eggs in the flesh of the olive fruit. Each female lays 200 to 250 eggs

the flies before they lay their eggs. For this purpose, it is essential, according to Trappmann (40), to carry out the bait sprays at intervals of two weeks. The Berlese method was employed on a large scale after the first world war in Italy and Greece. In 1921, the Berlese method was used by Isaakidès to treat 4,000,000* trees in Corfu, and in Italy a government decree was issued on February 21, 1922 enforcing the application of the Berlese method in the threatened districts of Central Italy (39). In 1923, the Spanish government promoted an olive fruit fly congress in Madrid, at which consultations were held on the organization of joint and uniform control operations. The resolution passed by

* This figure quoted by Trappmann (39) is, according to Triantaphyllidis (40a), far too high, for even today the number of olive trees in Corfu ist still approx. 1/4 less than given above.

this congress recommended member governments to enact legislation enforcing simultaneous controls of the olive fruit fly by means of the Berlese method in the hazarded areas. It was also recommended to establish cooperative associations, along the same lines as the so-called "olive banks" in Greece, in order to have funds available at all times for financing the control operations (39). The Berlese method remained the only recognized practice for controlling *Dacus* in the whole of the Mediterranean zone until after the second world war. Peretz and Plaut (33) report on trial controls of olive fruit fly carried out from 1940 to 1944 in Israel by means of the Greek modification of the Berlese method. By this method, parts of boughs on all trees growing in a certain

Fig. 2: The eggs hatch in summer after 2 to 3 days; larval development lasts 14 days. The larvae feed on fruit flesh, and pupate in the fruit

region are each treated with 500 ccs of bait liquid consisting of 10—15 % sugar cane or beet molasses, 0·25 % sodium arsenate and 0·5 % ammonium sulphate in aqueous solution (1). The spraying dates are fixed by suspending from a number of trees, trap bottles filled with ammonium sulphate (attractant) in order to check the flight. The bottles are examined for trapped flies every five days, and the next control date is fixed according to the numbers of flies caught. Gómez Clemente and Bellod (14) report on many years of tests (1942 to 1949) carried out in Spain, where the use of trap bottles filled with ammonium sulphate, the typical *Trypetidae* bait (42), constituted the only control method. In this paper, a report is given on comparison tests in which the trap bottle method proved just as successful as the bait spray method. In regions of high relative humidity, the trap bottle method is not a success (43), as demonstrated by the tests of Kalopissis (17 b).

Control techniques have changed in the last ten years. The chlorinated hydrocarbons and organophosphorus compounds have now become established also for controlling olive fruit fly; instead of using bait sprays which were successful only when used on a large scale and simultaneously by all, the trees and fruits are now copiously wetted by cover spray solution. Logothetis (21, 22) several years ago fully investigated the advantages and disadvantages of both spraying methods in olive growing, bait spraying and cover spraying. The advantage of cover spraying by means of organophosphorus compounds lies in the simultaneous effect given against adults, larvae and eggs with only 1 to 2 applications per year as against at least 6 applications for the Berlese method which gives exclusive and inadequate control of adults.

Initially dichlorodiphenyltrichloroethane, HCH, dieldrin and other chlorinated hydrocarbons were used for cover spraying but very soon the importance of E 605® forte (ethyl-parathion) for this method was recognized. According to Ayoutantis et al (7), the first tests with parathion against olive fruit fly were carried out by Martelli in 1949 in Italy; these were followed by numerous other tests. The application of chlorinated hydrocarbons for controlling olive fruit fly by the cover spray method did not always produce satisfactory results. As Logothetis (22) points out, the main feature of dichlorodiphenyltrichloroethane is its effect on adults, for which at least 2 to 3 applications are needed. Martin (22a) reports on tests carried out in Algeria, in which dust applications of dichlorodiphenyltrichloroethane at intervals of two weeks reduced infestation by only about fifty percent, although this treatment gave much better results in Spain. The author believes that this difference in the results obtained may be accounted for by greater olive fruit fly resistance to dichlorodiphenyltrichloroethane at temperatures above 30° C. Tominić (38) reports that aircraft applications of dichlorodiphenyltrichloroethane at high temperatures gave poor results. Kusmić (19) mentions tests in which dichlorodiphenyltrichloroethane was atomized 7 times in one case and dusted 13 times in another case in order to give effective control of *Dacus*. Melis (23) also reports on unsatisfactory results obtained with dichlorodiphenyltrichloroethane, HCH and toxaphene. Russo (35) is, therefore, of the opinion that the application of dichlorodiphenyltrichloroethane, HCH and dieldrin should be restricted to preventive sprayings in June for controlling adults (before egg-laying). Avidor, Mohrer and Harpaz (5) obtained good results with dieldrin against larvae and adults. But at the 2nd and 3rd FAO Meetings on the Control of the Olive Fly, held in Athens in 1955 and in Florence in 1957, only little time was devoted to the subject of chlorinated hydrocarbons (43, 44). Large-scale control operations conducted against the olive fruit fly with parathion in different countries produced better results, and especially a longer residual effect, which is a most important factor considering that olive fruit fly control is also a question of costs. Boselli (9) mentions that the olive yields in Sardinia are so low that even a control by means of the Berlese method is uneconomical. The main attention of research workers testing new products is, therefore, concentrated on the possibility of a long residual effect being given against larval stages, accompanied by a simultaneous action against adults. A further aspect that is given consideration is the demand for a non-toxic insecticide in order to avoid any toxic residues in the olive oil.

Extremely good results at low costs were obtained with E 605® forte and other parathion preparations by Ayoutantis et al (7, 8), Melis (23, 24, 25),

Russo and Santoro (36) and Mijušković and Mirčetić (26) — to mention but a few authors — so that in view of this, the authorities made every endeavour to keep developments under their control for toxicological reasons. In Greece, for example, exact safety periods were laid down for parathion (and also for other products) — that is the period that must elapse between final application of the product and harvesting of the crop — and the concentration for application of parathion was restricted to 0.025 % active ingredient. In Italy, an attempt was made to minimize the persistence of parathion in olive oil, by adding copper sulphate to the parathion spray solution. Russo (35) reports that the residues in olive oil were smaller for copper-parathion combinations

Fig. 3: The punctures made by the olive fruit fly are perceptible as dark spots on the olives, and render the fruit unfit for eating. The tunnels bored by the larvae are filled with oil. Top: Exit holes of the fly

than for parathion itself; according to Melis (25) the copper-parathion combinations did not lead to smaller residues. Russo and Santoro (36) state that copper sulphate increased the effect of parathion, and reduced the occurrence of the egg parasite *Prolasioptera berlesiana*, by virtue of the effect of copper sulphate against *Macrophoma dalmatica; P. berlesiana* always occurs in association with *M. dalmatica*. According to Pietri-Tonelli and Barontini (33 a) it was found in laboratory tests that an addition of copper had no influence on the effect and the speed of action of parathion.

In recent years, numerous other preparations have also been tested for their effect on olive fruit flies, especially with the object of discovering products with a low order of toxicity. Rupp (34), Ciampolini (11), Melis (24), Avidov, Moore & Harpaz (6), Ayoutantis et al (8) report on the use of

diazinon, and Orphanidis and Tsakmakis (32) on plant injuries caused by application of malathion. Extensive tests were carried out by Orphanidis and co-workers (30) in 1957; they used parathion, diazinon, Nexion®, Ekatin®, Rogor®, Ciba 570 and DIPTEREX®. Furthermore, Orphanidis and co-workers (27) tested the systemic action of phosphamidon and thiometon, and Orphanidis and Adam (31) investigated the problem of residues for numerous active ingredients.

Since 1952 in Morocco and since 1955 in Greece (43, 44), large-scale control operations have been in progress, primarily in table olive plantations with preventive sprays being carried out in spring against *Dacus oleae* imagines and all stages of *Prays oleellus* Fabr. The olive kernel borer which occurs in 3 generations every year, causing considerable damage in certain areas, and which was not affected by previous control methods employed against olive fruit fly — such as the Berlese method or curative cover sprays applied in summer — is most effectively controlled in the first half of the blossom period when the larvae of the second brood attack the buds and blooms (2, 44). The practice of controlling olive fruit fly in spring is also supported by the observation that in the spring months the adults are to be found solely on the olive trees (44), and can thus be effectively controlled. A further advantage of spraying in spring is that at this time of the year, the question of residues need not be given consideration, a factor that is especially important in the cultivation of table olives. Another point of great significance with respect to table olives is that the punctures made by the olive fruit fly develop into scars in the form of dark spots, thus resulting in a considerable lowering of quality; if only because of this, it is essential that controls of *Dacus* are carried out early enough to prevent egg-laying. It was this aspect which was foremost in the minds of research workers in Greece (44) when they carried out the following treatments using Parathion (0·2 % active ingredient) in 2 to 3 spray applications, from 1955 to 1957: in 1955 treatment of 25,000 trees in Rovies, in 1956 35,000 trees in Rovies, 350,000 trees in Corfu, and in 1957 a total of 1,380,000 trees. The results of these tests revealed that by carrying out a control in spring, the *Dacus* population can be suppressed until September (44). In olive plantations cultivated for oil production, the spray applications still required should then be carried out according to severity of infestation, while the maturing black or blackish-blue table olives no longer hold any attraction for the olive fruit fly to lay its eggs on them (passive resistance), and thus escape late infestation. Green table olives, on the other hand, should be treated at this time with phosphate bait sprays which, unfortunately, like the trap bottle method, do not always give satisfactory results in Greece due to the high relative humidity (43, 44, 29). Consideration should be given in this respect to experience gained by Steiner (37) and to that accumulated in Spain (44) in the control of *Dacus dorsalis, Dacus oleae* and *Ceratitis capitata.*

With the object of furthering the biological control of olive fruit fly, the International Commission for Biological Control (CILB) formed in 1956 a working group to deal with the biological control of *Dacus oleae* and *Ceratitis capitata* (44). Hennig (15) reports that Silvestri (in Italy) and Nonell Comas (in Spain) did not have much success in the past when they introduced *Opuis concolor* for biological control of *Dacus.* Russo and Santoro (36), on the other hand, observed considerable parasitization of larvae by *Dinarmus dacicida, Eurytoma rosae, Pnignalio mediterranens (= Eulophus longulus)* and

Eupelmus urozonus in experiments they conducted in 1954 in Italy; eggs, too, were considerably parasitized by *Prolasioptera berlesiana* already mentioned in another connection as occurring in association with *Macrophoma dalmatica*. Isaakidès (16) reports on severe occurrence of the ichneumon fly *Eupelmus urozonus* as a parasite of *Dacus* in Greece and Crete; 80 % of the larval population was observed to be parasitized by this chalcid. The author attributes the great abundance of *Eupelmus urozonus* to the presence of suitable host insects. In regions where the olive tree grows together with St.-John's-bread *(Ceratonia siliqua)* and *Inula viscosa, Eupelmus urozonus* finds host insects all the year round for maintaining its population, namely *Eumarchalia gennadii* as a pest of St.-John's-bread and *Myropites limbardae* on *Inula viscosa,* so that when olive fruit fly infestation begins, a perceptible parasitization of this pest by ichneumon flies already present on other hosts can take place. Ferrière and Delucchi (13) report that chalcids occur generally as parasites of *Dacus* in the Mediterranean zone. It is however, pointed out that ichneumon fly occurrence leads to rapid extermination of the host insects only in the rarest of cases. According to Trappmann (40), "we find, on the contrary, that among endoparasitic insects there is a highly developed parasitism, in that the parasite exploits its host for as long as possible by not harming the host's vital organs (nervous system, digestive organs) in oder to live in the host's body and obtain nourishment from it until the parasite pupates". Therefore, in the case of the olive, parasitization does not have the direct effect of preventing damage to the individual fruit but may lead under certain circumstances to a reduction of the subsequent infestation as viewed from the aspect of the total population. Past experience has shown that the practice of chemical control cannot be abandoned in olive growing; on the contrary, it is absolutely essential for preventing damage by olive fruit fly infestation.

II. Field trials in Crete*

Formulation of problem

The present situation in the control of olive fruit fly requires the provision of products that feature a rapid initial effect and a long-lasting residual action against larval stages, also a good effect against adults, accompanied by a low order of mammalian toxicity. In addition to the action against adult flies, great importance must be attached to the residual effect against larvae because it is extremely essential that very persistent products are used in olive orchards for economical and technical reasons. Due to the trackless nature of the country in the olive growing regions in the hills, every single spray application incurs such high expenditure for labour and working hours that an attempt must be made to give the crops adequate treatment and protection with as few aimed sprays as possible. In our large-scale field experiments conducted from September 1 to November 16, 1958 in the district of Chania on the island of Crete, the prime object in mind was to test the residual action of LEBAYCID as an emulsion** and a wettable powder***. Apart from testing the persistence

* I am especially indebted to Director J. Raftopoulos, head of the Phytopathological Division of the Greek Ministry of Agriculture in Athens, Director N. Micheliodakis, head of the regional agricultural administration in Chania, Crete, and N. Psilakis, agr. dipl., agricultural adviser in Kolybari, Crete, for the invaluable and kind assistance they rendered us while carrying out these tests.

** Test No. 4895

*** Test No. 4894, 4970.

of LEBAYCID against larvae at different concentrations, we were also interested in establishing the speed of the initial action produced against the different larval instars and the effect given by LEBAYCID against adults.

LEBAYCID (S 1752, Bayer 29493) is an insecticide with a low order of toxicity, synthetized by Schrader (36a, 36b) at the crop protection research laboratories of Farbenfabriken Bayer AG in Elberfeld. Unterstenhöfer (41) has published a paper including details of the laboratory and greenhouse tests carried out against different agricultural pests (special mention is made, inter alia, of the good effect against *Trypetidae)* at the Biological Institute of Farbenfabriken Bayer AG in Leverkusen; Jung (17a) reports on laboratory tests against pests of public health importance.

Apart from LEBAYCID, our olive fruit fly tests included FOLIDOL E 605® forte and FOLIDOL M 50. Ethyl parathion is the lowest-priced product at present in use for controlling olive fruit fly although it has the disadvantage of possessing a relatively high order of mammalian toxicity, whereas FOLIDOL M 50 (methyl parathion) is less toxic. Methyl parathion has not yet been used to our knowledge for controlling *Dacus oleae.* Further products tested included diazinon, thiometon, METASYSTOX® and several other test preparations.

For the purpose of carrying out residue studies, olives were sampled in all tests and pressed, being primarily taken from the LEBAYCID plots; the results of these studies will be published in a separate paper.

Fig. 4: Due to the special needs of the olive tree for a well-balanced climate free of frost conditions, cultivation of olives is restricted to the coastal provinces and islands of the Mediterranean countries. Crete is one of the chief growing areas by virtue of its favourable climate for olive tree growth (according to Allbaugh and Soule [1])

In planning these tests, the foremost principle was to carry them out in a typical olive growing region where infestation conditions were as favourable as possible; this is the case in the hilly parts and in the plain around Chania in north-west Crete. The island of Crete is one of the world's oldest olive growing regions. Records show that olive growing was practised in Crete as far back as 2000 to 3000 B. C. According to Allbaugh and Soule (1), Crete today produces the highest oil yields per tree in the whole of the Medi-

terranean zone, and the total yield obtained from the island's 13,000,000* trees amounts to 27 % of the Greek olive oil production (1). Olives are grown in the hilly districts up to heights of 600 metres, and in fact several varieties thrive at 800 metres a. s. l. Olive orchards comprise an area amounting to 16 % of the acreage of agricultural land in Crete. It is understandable that the olive fruit fly has always been a native insect pest of this island; before the cover spray method, using parathion, was introduced some ten years ago, considerable sums had been expended since 1920 for control operations by means of the Berlese method. According to Allbaugh and Soule (1), the costs of the Berlese control in 1947 in Crete amounted to 160 000 dollars, and in 1948 to 80 000 dollars. Especially interesting information in this connection is provided by the studies also carried out by Allbaugh and Soule (1) to establish the extent of olive fruit fly damage in Crete. The farmers estimate the average damage at 44 %, and the estimates of the agricultural offices show figures varying between 15 and 80 % for the different years according to severity of infestation. When it is considered that in Crete 97 % of the farmers grow olives — even though some of them do so solely for their own consumption — these figures again emphasize the great importance of olive fruit fly control.

Implementation and evaluation of tests

All the tests were carried out in the area around Chania, some near the coast and the others in the hilly districts (at heights of up to 400 to 500 metres). Motor-powered sprayers were used in all the tests; as far as possible, medium-sized trees severely infested by new colonies were selected for the tests. In the first tests, the products were sprayed at high concentrations, and in the others carried out after September 15, low concentrations were used in compliance with the recommendations of the Greek Ministry of Agriculture regarding the concentrations to be applied for the different commercial products. Details of the tests are given below. The concentrations quoted refer to active ingredient.

Testing the effect against larvae

Test No. 1: Spraying on Sept. 1, 1958 at Perivolia,
Tsunati olive variety, 2 trees per product resp. concentration
Sprayer type: Platz Patria Z

Weather: sunny, min. temp. 20 ° C, max. temp. 30 ° C, relative humidity 74 %

Products:	
DIPTEREX® Emulsion	0·1 %
DIPTEREX Emulsion	0·15 %
LEBAYCID Emulsion	0·06 %
LEBAYCID Emulsion	0·1 %
LEBAYCID Wettable Powder	0·06 %
LEBAYCID Wettable Powder	0·1 %
FOLIDOL M 50	0·05 %
FOLIDOL E 605 forte	0·05 %
4961	0·06 %
4891a	0·06 %
Diazinon	0·06 %
Untreated check	

Test No. 2: Spraying on Sept. 2, 1958 at Perivolia,
Tsunati olive variety, 2 trees per product resp. concentration
Sprayer type: Platz Patria Z

* According to Triantaphyllidis (40a), the stock of olive trees in Crete amounted to 14,317,180 in 1959.

Weather: sunny, min. temp. 21 0 C, max. temp. 30·4 0 C, relative humidity 70 %.

Products:		
	FOLIDOL E 605 forte	0·05 %
	FOLIDOL M 50	0·05 %
	FOLIDOL M 50	0·025 %
	LEBAYCID Emulsion	0·06 %
	LEBAYCID Emulsion	0·1 %
	LEBAYCID Emulsion	0·15 %
	DIPTEREX Emulsion	0·1 %
	DIPTEREX Emulsion	0·15 %
	Untreated check	

Test No. 3: Spraying on Sept. 8, 1958 at Perivolia,
Tsunati olive variety, 5 trees per product resp. concentration
Sprayer type: Platz Patria Z

Weather: sunny, min. temp. 19·4 0 C, max. temp. 26·4 0 C, relative humidity 70 %.

Products:		
	DIPTEREX Emulsion	0·1 %
	DIPTEREX Emulsion	0·15 %
	LEBAYCID Emulsion	0·06 %
	LEBAYCID Emulsion	0·1 %
	FOLIDOL M 50	0·05 %
	FOLIDOL E 605 forte	0·05 %
	Diazinon	0·06 %
	Untreated check	

Test No. 4: Spraying on Sept. 9, 1958 at Perivolia
Tsunati olive variety, 4 to 6 trees per product resp. concentration
Sprayer type: Platz Patria Z

Weather: sunny, min. temp. 17·8 0 C, max. temp. 27 0 C, relative humidity 66 %.

Products:		
	LEBAYCID Emulsion	0·06 %
	LEBAYCID Emulsion	0·12 %
	LEBAYCID Emulsion	0·1 %
	LEBAYCID Wettable Powder	0·06 %
	LEBAYCID Wettable Powder	0·12 %
	DIPTEREX Emulsion	0·12 %
	4891a	0·06 %
	FOLIDOL E 605 forte	0·06 %
	FOLIDOL M 50	0·06 %
	Untreated check	

Test No. 5: Spraying on Sept. 17, 1958 at Palea Rumata,
2 olive varieties Tsunati and Lianolia, 10 trees per product resp. concentration, 5 trees of each olive variety
Sprayer type: Platz Patria Z

Weather: sunny, min. temp. 17·2 0 C, max. temp. 26·6 0 C, relative humidity 80 %.

Products:		
	LEBAYCID Emulsion	0·06 %
	LEBAYCID Emulsion	0·03 %
	FOLIDOL M 50	0·03 %
	Diazinon	0·03 %
	FOLIDOL E 605 forte	0·03 %
	Untreated check	

Test No. 6: Spraying on Sept. 30, 1958 at Glossa,
Lianolia olive variety, 5 trees per product resp. concentration
Sprayer type: Platz Patria Z

Weather: sunny, min. temp. 16·6 0 C., max. temp. 23·8 0 C, relative humidity 69 %.

Products:	DIPTEREX Emulsion	0·03 %
	LEBAYCID Emulsion	0·03 %
	LEBAYCID Emulsion	0·015 %
	4891a	0·03 %
	METASYSTOX	0·03 %
	Diazinon	0·03 %
	FOLIDOL E 605 forte	0·03 %
	FOLIDOL M 50	0·03 %
	Thiometon	0·03 %
	LEBAYCID Wettable Powder	0·03 %
	LEBAYCID Wettable Powder	0·015 %
	4961	0·03 %
	4961	0·015 %
	Untreated check	

Test No. 7: Spraying on Oct. 12, 1958 at Chania,
Tsunati olive variety, 120 trees per product resp. concentration
Sprayer type: Platz Patria Z
Weather: sunny, min. temp. 16·2 ° C, max. temp. 22 ° C, relative humidity 76 %.
Product: LEBAYCID Emulsion 0·3 %

Test No. 8: Spraying on Oct. 15, 1958 at Chania,
Tsunati olive variety, 120 trees per product resp. concentration
Sprayer type: Platz Patria Z
Weather: sunny, min. temp. 18·7 ° C, max. temp 24·6 ° C, relative humidity 73 %.
Product: LEBAYCID Wettable Powder 0·03 %

The evaluation of the tests to establish the residual effect of the products, was carried out by means of the method customary in Greece; olives were sampled at intervals of 5 to 10 days (50 to 100 olives per product), and examined for olive fruit fly infestation according to the following plan:

No. of punctures	No. of eggs	L_1				L_2		L_3		Pupae		Exit holes*	
		Dead		Living									
		with galleries	without galleries	with galleries	without galleries	D	L	D	L	D	L	No. of fruits	No. of fruits

* Exit holes of flies that emerged from pupae in the fruit; in autumn, these holes were the exits of L_3 that left the fruit to pupate.

In this way, an exact analysis of the population is made in each evaluation so that as the residual effect lessens new infestation can be well observed. Infestation increased in all tests as the residual effect lessened, thus constituting a favourable development for sampling and evaluation.

An evaluation of the initial effect against older larval stages was carried out in test No. 6 (Glossa); after periods of 1, 2, 3 and 5 days, 50 and 35 L_2 and L_3, respectively, were checked for percentage of mortality per product and concentration, respectively.

Testing the effect against flies

These experiments were limited to testing of LEBAYCID emulsion in comparison with a commercial preparation*; the products were applied as cover sprays (no bait sprays) at 0·03 % resp. 0·015 % active ingredient.

Test No. 1: Spraying on October 12, 1958 at Chania
Tsunati olive variety, 2 trees per product
Sprayer type Platz Patria Z
Size of trap sheet per tree: 60 square metres

Weather: sunny, min temp. 16.2 ° C, max. temp. 22 ° C, relative humidity 76 %, wind force 1 (Beaufort)

Products:

LEBAYCID Emulsion	0.03 %
Comparison product*	0.03 %
Untreated check	
Duration of observation: 21 days	

Test No. 2: Spraying on Oct. 14, 1958, morning, at Kares,
Lianolia olive variety, 2 trees per product resp. concentration
Sprayer type Platz Patria Z
Size of trap sheet per tree: 40—50 square metres

Weather: sunny, min. temp. 13.7 ° C, max. temp. 25.6 ° C, relative humidity 64 %, wind force 0 (Beaufort)

Products:

LEBAYCID Emulsion	0.03 %
Comparison product*	0.03 %
FOLIDOL M 50	0.03 %
4961	
Untreated check	
Duration of observation: 9 days	

Test No. 3: Spraying on Oct. 27, 1958 at 5 p. m., at Chania,
Tsunati olive variety, 3 trees per product resp. concentration
Sprayer type Platz Patria Z

Weather: sunny, min. temp. 15.4 ° C, max. temp. 20.4 ° C, relative humidity 77 %, wind force 0 (Beaufort)

Products:

LEBAYCID Emulsion	0.03 %
Comparison product*	0.03 %
Untreated check	
Duration of observation: 10 days	

Test No. 4: Spraying on Nov. 1, 1958, at 5 p. m., at Chania,
Tsunati olive variety, 2 trees per product resp. concentration
Sprayer type Platz Patria Z
Size of trap sheet per tree: 60 square metres

Weather: sunny, min. temp. 15 ° C, max. temp. 19.2 ° C, relative humidity 73 %, wind force 0 (Beaufort)

Products:

LEBAYCID Emulsion	0.03 %
Comparison product*	0.03 %
Untreated check	
Duration of observation: 5 days	

Test No. 5: Spraying on November 6, 1958, at 5 p. m. at Chania,
Tsunati olive variety, 4 trees per product resp. concentration
Sprayer type Platz Patria Z
Size of trap sheet per tree: 60 square metres.

Weather: sunny, at times cloudy, min. temp. 16.7 ° C, max. temp. 23 ° C, relative humidity 79 %, wind force 0 (Beaufort)

Products:

LEBAYCID Emulsion	0.03 %	2 trees from Test No. 3 and 2 trees from Test No. 4
Comparison product*	0.03 %	
Untreated check		
Duration of observation: 1 day (check on residual effect given by second spray after 5 and 10 days).		

* N-monomethylamide of 0,0-dimethyldithiophosphoryl acetic acid.

The size of the trap sheets spread out under each tree was usually 60 square metres as stated above; these trap sheets were chiefly laid out on the south side of the trees. To determine the rate of mortality the flies were collected from the sheets in the morning (night knock-down) and evening (day knock-down), and counted. In order to ensure that the trap sheet tests would proceed without any outside interference, they were carried out near to the houses of reliable farmers, and a nightwatchman was posted to guard the trap sheets from dusk to dawn.

Fly infestation was considerable in tests 1, 3, 4 and 5 so that it was primarily possible to check the residual effect against adults.

Fig. 5: It is most important to use insecticides of low toxicity for controlling olive fruit fly because olives are a staple food in numerous countries. Therefore, in our field tests olive samples were pressed at the same time for residue analyses

Results

Implementation of the tests was rendered difficult in the first half of September due to the fact that the heat-wave in August had greatly decimated the *Dacus* population. Table 1 shows that in August maximum temperatures of 40 ° C were recorded, accompanied by very low relative humidity. Furthermore, there was very little rainfall, which had a generally unfavourable effect on the olive yield.

The evaluation of the tests was also rendered difficult in September due to severe occurrence of parasites, although no killed parasite larvae were found in the counts for any of the tested products.

Initial effect

In order to test the initial effect of the different products, they were sprayed at equal concentrations of active ingredient in test No. 6 carried out

Table 1: Climatic data recorded at the meteorological station, Chania, Crete, over four years*

Year	Month											
	Jan.	Feb.	March	April	May	June	July	Aug.	Sept.	Oct.	Nov.	Dec.
						Mean temps. °C						
1955	13·3	15·1	13·8	13·3	21·1	24·5	27·0	25·4	23·2	21·4	17·2	14·2
1956	12·4	11·5	10·9	16·4	19·4	23·8	26·6	25·9	22·5	18·5	17·9	12·1
1957	10·7	13·1	12·1	15·2	18·5	24·5	25·3	26·3	23·4	20·1	16·4	12·5
1958	11·6	13·1	14·2	16·0	21·1	24·4	26·1	26·4	22·1	19·2	16·7	14·4
						Max. temps. °C						
1955	21·2	28·0	27·2	31·8	34·6	35·4	39·6	32·0	31·0	32·0	31·0	23·0
1956	21·4	22·2	20·0	34·0	31·9	35·2	41·0	35·0	34·0	31·0	29·2	24·6
1957	20·0	26·0	24·0	29·3	30·0	39·0	34·2	37·0	33·2	30·0	28·4	26·4
1958	20·4	25·2	26·0	26·0	34·8	36·2	36·0	40·0	37·4	31·0	25·0	22·9
						Min temps. °C						
1955	8·5	7·8	3·8	7·8	12·0	17·0	20·6	19·6	17·0	15·2	9·0	7·8
1956	6·4	0·4	4·0	6·4	10·4	16·5	18·4	19·2	13·2	11·0	9·8	5·0
1957	5·2	4·6	3·6	8·0	11·2	16·2	16·8	19·5	16·6	14·5	11·5	4·0
1958	5·4	6·1	7·8	8·0	10·4	15·8	19·0	19·6	16·0	12·6	11·0	7·8
						Mean relative humidity %						
1955	79	61	65	67	59	59	56	61	68	68	71	74
1956	71	71	70	59	60	58	52	59	63	64	69	69
1957	74	64	69	69	72	55	56	61	67	76	76	71
1958	72	70	67	65	61	55	55	54	69	68	79	76
						Max. relative humidity %						
1955	95	95	95	95	96	94	91	65	95	94	93	95
1956	93	100	94	91	92	92	91	91	90	93	92	95
1957	95	93	95	93	96	92	84	90	92	96	93	95
1958	93	93	93	94	89	86	83	81	91	90	93	93
						Min. relative humidity %						
1955	41	13	16	22	19	22	15	31	30	24	26	38
1956	40	35	34	14	25	23	13	26	36	17	34	36
1957	41	15	25	27	34	25	27	19	28	46	36	35
1958	42	28	23	29	12	20	16	24	36	35	56	42
						Rainfall mm						
1955	66·0	22·9	81·3	25·1	3·8	—	—	2·3	106·7	47·7	133·5	124·1
1956	74·3	288·6	94·7	—	18·7	—	—	—	6·2	32·0	6·3	87·2
1957	315·3	11·6	127·0	33·3	27·1	4·6	0·4	—	19·1	129·4	46·6	115·9
1958	93·6	8·8	43·9	16·6	3·7	4·2	0·2	—	45·1	55·8	82·5	168·8
						Annual rainfall mm						
1955	613·4											
1956	608·0											
1957	830·3											
1958	532·2											

* These data were kindly provided by Mr. Drosoulakis, head of the meteorological station at Chania. Cf data of Allbaugh and Soule (1)

Table 2: Test No. 6 (Glossa); initial effect against older larval stages

Spray application on Sept. 9, 1958	Conc. of active ingredient in %	Percent of dead, injured and living larvae counted after											
		1 day			2 days			3 days			5 days		
		dead	injured	living	dead	injured	living	dead	injured	living	dead	injured	living
DIPTEREX-Emulsion	0·03	40	27	33	80	12	8	100	—	—	93	7	—
LEBAYCID-Emulsion	0·03	92	8	—	92	8	—	100	—	—	100	—	—
LEBAYCID-Emulsion	0·015	80	12	8	84	16	—	93	7	—	100	—	—
4891 a	0·03	10	—	90	53	13	34	73	27	—	86	7	7
METASYSTOX	0·03	20	10	70	53	7	40	66	7	27	73	7	20
Diazinon	0·03	44	24	32	60	27	13	67	13	20	86	—	14
FOLIDOL E 605 forte	0·03	56	4	40	60	40	—	86	7	7	100	—	—
FOLIDOL M 50	0·03	80	13	7	100	—	—	93	7	—	100	—	—
Thiometon	0·03	24	16	60	53	7	40	66	20	14	80	7	13
LEBAYCID Wettable Powder	0·03	36	56	8	100	—	—	100	—	—	100	—	—
4961	0·03	70	20	10	100	—	—	100	—	—	100	—	—
4961	0·015	47	33	20	100	—	—	93	7	—	100	—	—
LEBAYCID Wettable Powder	0·015	32	40	28	93	7	—	100	—	—	100	—	—
Untreated	—	4	8*	88	—	7*	93	—	20*	80	—	13*	87

* Pupae

on September 30, 1958 at Glossa. Small trees were selected for this test, which were easy to spray, which had a good coverage of fruit, were severely infested by olive fruit fly and colonized by few parasites. 1, 2, 3 and 5 days after the application, 50 resp. 25 larvae (stages 2 and 3 only) were examined per product resp. concentration for living, dead and injured individuals. It is obvious from the results in Table 2 that the various products differ considerably in their initial effect.

The best initial effect at the same concentration of active ingredient, namely 0·03 % and 0·015 %, was given by LEBAYCID (Emulsion and Wettable Powder), followed by 4961, FOLIDOL M 50 and DIPTEREX; practically 100 % mortality of the older larval stages was obtained after one to two days. The

Fig. 6: Young plantations are especially suitable for test work regarding exactness of spraying technique and facilitation of infestation evaluation (Test 6)

rate of mortality given after 24 hours by the emulsion of LEBAYCID is better than the wettable powder formulation of this product. No appreciable differences in the initial effect were established between the active ingredient concentrations 0·03 % and 0·015 % for any of the products tested at two strengths (LEBAYCID Emulsion and Wettable Powder and 4961). FOLIDOL E 605 forte (ethyl parathion) did not give 100 % mortality of L_2 and L_3 until after five days had elapsed, in contrast to FOLIDOL M 50 (methyl parathion) which had a better initial action. There was still a considerable percentage of living larvae after five days on trees treated with 4891a, diazinon, Thiometon and METASYSTOX.

The initial effect given against older larval stages is of secondary importance in comparison with the residual effect because timely sprays are carried out before the larvae have caused any damage in the fruits, in other words,

sprays are applied at a time when only eggs and L_1 are present. However, as growers do not always keep to this schedule, a good initial effect of the products against older larval stages is also of importance, for the fruits in which L_2 and L_3 are already tunneling at the time of the application in most cases recover from feeding damage and ripen as a result of the immediate eradication of the larvae.

In the other tests, the findings corresponded to these results with respect to the initial action. In immersion tests, in which two batches of fifty olives infested by L_2 and L_3, respectively, were treated with active ingredient concentrations of 0·03 % and 0·01 % and evaluated after 10 and 24 hours, respectively, the same order of mortality was obtained for the different products.

Residual effect against continuous infestation

Several tests were thoroughly evaluated for the purpose of determining the residual effect. Six large spray tests were carried out because it could not be said in advance whether the infestation at the different test sites would be adequate over the whole of the observation period, in other words until the residual effect had faded for all products.

As the eggs hatch after a few days, it is essential that new eggs are laid almost interruptedly in order to test the residual action. In the evaluation of the tests, an exact analysis of the population is, therefore, carried out at each count made at intervals of 5 to 10 days, in order to establish the number of punctures and eggs, living and dead L_1, L_2, L_3 and pupae, and also the number of exit holes. In this manner, lessening of the residual effect can be established for the different products from the abundance of live L_1 and, in the event of these not being eradicated, from the presence of live L_2 and L_3 at the next count.

It is evident from the untreated check of test No. 6 (Glossa) in Table 3, that live larvae of all stages were found at all counts throughout the whole of the observation period lasting fifty days, which means that infestation was continuous. The results in Table 2 for the experiments carried out to test the initial action, show that the older larval stages had also been eradicated after five days, with the exception of a few products. According to table 3, a few living larvae were found after 10 days only following the spray application of DIPTEREX also after fifteen days, whereby the later counts show that there is the gradual increase in the number of live larvae also among the older stages. New infestation sets in at a steady rate following application of the other products. In the case of 4891a and 4961, live larvae are found after 20 to 30 days, for Thiometon, METASYSTOX, FOLIDOL M 50 and FOLIDOL E 605 forte after 30 days, for Diazinon after 35 days, for LEBAYCID Emulsion and Wettable Powder after 40 to 45 days. It should, however, be mentioned that for LEBAYCID (Emulsion and Wettable Powder) only very few older larval stages were found at the last count; sampling was carried out after 47 days but the fruits were not examined until after 54 days. This large interval is left between sampling and examination in autumn for the simple reason that larval development progresses more slowly at the dropping temperatures. When the samples are left to stand at room temperature for a few days, the larvae develop more quickly; this makes the examination much easier and also increases the accuracy of the results, for the young larvae (L_1

Table 3: Test No. 6 (Glossa). Residual effect for low active ingredient concentrations (number of live larvae in batches of 50 olives).

Spraying on Sept. 30, 1958	Sampling: 10. 10. (10 days) Examination: 11. 10. (11 days)					13. 10. (13 days) 15. 10. (15 days)					21. 10. (21 days) 22. 10. (22 days)					26. 10. (26 days) 28. 10. (28 days)					1. 11. (32 days) 7. 11. (38 days)					6. 11. (37 days) 11. 11. (42 days)					12. 11. (43 days) 17. 11. (48 days)					16. 11. (47 days) 23. 11. (54 days)				
	active ingredient %	L_1		L_2	L_3	S	L_1		L_2	L_3	S	L_1		L_2	L_3	S	L_1		L_2	L_3	S	L_1		L_2	L_3	S	L_1		L_2	L_3	S	L_1		L_2	L_3	S	L_1		L_2	L_3	S
		mM	oM				mM	oM				mM	oM				mM	oM				mM	oM				mM	oM				mM	oM				mM	oM			
DIPTEREX Emulsion	0·03	—	—	2		1	—	1	—	—	—	—	—	1	—	2	7	1	2	—		5	2	5	1	—	6	3	4	2	10	—	—	5	19	27	4	—	7	18	14
LEBAYCID Emulsion	0·03	—	—	—	—	2	—	—	—	—	1	—	—	—	—		—	—	—	—	—	—	—	—	—	—	—	3	—	—	—	21	14	—	—	—	18	7	—	1	1
LEBAYCID Emulsion	0·015	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	1	10	—	—	—	15	8	—	—	—	20	14	1	—	—
4891 a	0·03	—	—	—	—	2	—	—	—	—	—	1	—	—	—	2	4	1	—	—	—	7	2	1	—	—	14	4	5	1	7	4	—	5	19	18	4	—	1	13	25
METASYSTOX	0·03	—	—	—	—	3	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	6	6	4	—	—	12	6	6	—	—	2	—	12	16	12	3	—	3	9	12
Diazinon	0·03	—	—	—	—	2	—	—	—	—	—	—	—	—	—	2	—	—	—	—	—	1	2	—	—	—	—	8	—	—	—	16	6	1	3	2	17	5	4	7	9
FOLIDOL E 605 forte	0·03	—	—	—	—	1	—	—	—	—	—	—	—	—	—	—	—	2	—	—	—	7	2	—	—	—	16	5	4	—	—	—	—	7	24	16	—	—	3	15	4
FOLIDOL M 50	0·03	—	—	—	—	—	—	—	—	—	—	—	—	—	—	2	—	—	—	—	—	10	1	—	—	—	12	8	1	—	—	1	—	7	24	6	1	—	9	17	19
Thiometon	0·03	—	—	—	—	6	—	1	—	—	—	—	—	—	—	1	—	—	—	—	—	8	3	1	—	—	18	6	1	—	—	28	—	2	4	10	6	—	3	24	17
LEBAYCID Wettable Powder	0·03	—	—	—	—	—	—	—	—	—	—	—	—	—	—	1	—	—	—	—	—	—	1	—	—	—	—	3	—	—	—	—	—	—	—	—	14	5	2	5	4
4961	0·03	—	—	—	—	—	—	—	—	—	1	—	—	—	—	3	1	5	—	—	—	10	2	1	—	1	15	2	5	4	—	10	—	10	16	7	1	—	4	14	5
4961	0·015	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	4	3	—	—	—	10	4	—	—	—	12	4	8	5	—	—	—	12	15	19	1	—	3	11	6
LEBAYCID Wettable Powder	0·015	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	2	—	—	—	1	3	—	—	—	8	5	—	—	2	18	5	2	1	1
Untreated		4	—	8	5	9	4	2	2		7	6	4	3	2	8	3	2	3	—	—	10	2	6	2	3	11	3	8	4	15	2	—	8	20	16	—	—	3	10	3

Explanation: The table includes only the absolute number of live stages found in fifty infested olives: L_1 with galleries, L_1 less galleries, L_2, L_3 and exit holes bored by flies which have emerged from pupae in the olives. The exit holes found on October 15 are those through which L_3 left the fruit in order to pupate in the ground. As pupae were found only in the untreated check at the first two counts, this column is not included in the table

mM = with tunnels oM = less tunnels

Table 4: Test No. 5 (Palea Rumata): Residual effect of LEBAYCID and other products in large and small olives (number of live larvae in batches of 50 olives)

Spraying on Sept. 17, 1958	Sampling: 17. 9. 1958 Examination: 19. 9. 1958 Initial infestation						27. 9. (10th day) 30. 9. (13th day)					17. 10. (30th day) 19. 10. (32nd day)					26. 10. (39th day) 30. 10. (43rd day)					1. 11. (45th day) 2. 11. (46th day)					5. 11. (49th day) 12. 11. (56th day)					13. 11. (56th day) 18. 11. (61st day)				
	active ingredient %		L_1		L_2	L_3	S	L_1		L_2	L_3	S	L_1		L_2	L_3	S	L_1		L_2	L_3	S	L_1		L_2	L_3	S	L_1		L_2	L_3	S	L_1		L_2	L_3	S
			mM	oM				mM	oM				mM	oM				mM	oM				mM	oM				mM	oM				mM	oM			
I. OLIVE VARIETY TSUNATI																																				
LEBAYCID Emulsion	0·06	top *	12	5	4	2	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	3	—	—	—	—	—	—	—	—	—	—	—	—
		bottom	17	0	4	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	12	6	4	1	—
LEBAYCID Emulsion	0·03	top	18	—	9	—	3	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	2	8	—	—	—	—	—	—	—	—	—	—	—	—
		bottom	30	—	15	6	—	—	—	—	—	—	—	—	—	—	—	—	2	—	—	—	—	2	—	—	—	4	5	—	—	—	4	1	2	5	1
FOLIDOL M 50	0·03	top	19	2	10	—	—	—	—	—	—	—	5	2	—	—	—	2	3	1	—	—	20	8	15	1	—	18	5	9	5	2				
		bottom	21	6	6	—	—	—	—	—	—	—	—	1	—	—	—	14	7	5	—	—	9	1	2	—	—	16	3	9	4	7	5	—	9	35	4
Diazinon	0·03	top	14	3	3	—	8	—	—	—	—	—	—	—	—	—	—	1	3	—	—	—	10	6	—	—	—	9	6	—	—	—				
		bottom	16	3	—	—	1	—	—	—	—	—	—	—	—	—	—	8	7	—	—	—	2	4	—	—	—	3	3	—	—	—	23	9	2	2	—
FOLIDOL E 605 forte	0·03	top	14	2	5	2	25	—	—	—	—	—	—	—	—	—	—	2	—	—	—	5	6	5	3	—	2	13	4	2	2	2				
		bottom	19	3	7	3	8	—	—	—	—	—	—	—	—	—	—																2	5	7	15	7
Untreated	—	top						5	—	4	8	7	—	—	4	3	6	6		3	1	6	4	—	4	5	12	2	3	3	2	16				
		bottom						2	—	3	3	2	5	—	4	2	11	15	4	7	5	9	6	—	5	3	4	4	—	6	6	8				
II. OLIVE VARIETY LIANOLIA																																				
LEBAYCID Emulsion	0·06	top	7	—	2	—	10	—	—		—	—	—	—	—	—	—	—	—	—	—	—	—	5	—	—	—	—	—	—	—	—				
		bottom	4	—	6	—	4	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—		3	—	—	—	12	4		2	—
LEBAYCID Emulsion	0·03	top	8	—	—	2	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—		3	—	—	—				
		bottom	12		2	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	3	1	—	—	—	9	5	1	2	—
FOLIDOL M 50	0·03	top	5	2	—	—	—	—	—	—	—	—	—	—	—	—	—	2	5	—	—	—	2	1	—	—	—	7	1	3	1	—				
		bottom	3	1	—	—	—	—	—	—	—	—	—	—	—	—	—	2	1	—	—	—	3	1	—	—	—	10	3	5	2	3	15	3	9	8	5
Diazinon	0·03	top	3	1	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	2	11	—	—	—	2	4	1	—	—				
		bottom	3	1	1	—	—	—	—	—	—	—	—	—	—	—	—	1	2	—	—	—	—	1	—	—	—	5	7	—	—	—	21	5	3	2	—
FOLIDOL E 605 forte	0·03	top	3	1	—	—	—	—	—	—	—	—	—	—	—	—	—	2	1	—	—	—	10	4	—	—	—	7	3	6	5	4				
		bottom	6	1	—	—	—	—	—	—	—	—	—	—	—	—	—	1	2	—	—	—	3	1	—	—	—	13	4	7	2	3	12	2	11	5	3
Untreated	—	top						5	—	2	5	6	3	—	2	1	4	5	1	3	6	10	6	—	3	2	5	3	2	5	3	6				
		bottom						4	1	3	2	3	2	—	1	—	7	8	2	6	1	7	5	—	4	1	3	15	3	4	2	4				

* top = olives sampled from tree top
bottom = olives from bottom outer branches
m M = with galleries
o M = less galleries

with and less tunnels) can only be detected with certainty by means of a binocular. It is also evident from the results given in Table 3 that there are slight differences in residual effect between the active ingredient concentrations 0·03 % and 0·015 %.

Table 4 gives the results of test No. 5 (carried out at Palea Rumata). In this test two olive varieties, a large fruit ("Tsunati") and a small fruit variety ("Lianolia"), were sprayed with LEBAYCID, FOLIDOL M 50, Diazinon and FOLIDOL E 605 forte all at an active ingredient concentration of 0·03 % (for LEBAYCID a second concentration of 0·06 % was also tested), when new

Fig. 7: Due to its small water requirements, the olive tree grows in high hilly country throughout the Mediterranean zone, thus enabling large areas to be utilized for agriculture. View of an olive orchard at Palea Rumata (Test No. 5) situated 500 metres a. s. l.

infestation commenced (initial infestation consisted only of eggs and L_1, but no L_2 or L_3). When the commencement of new infestation is assumed to indicate the termination of the effect, the following durations of residual effect are obtained:

	Active ingredient %	Olive variety Tsunati	Lianolia
LEBAYCID Emulsion	0·06	50 days	50 days
LEBAYCID Emulsion	0·03	45 days	45 days
FOLIDOL M 50	0·03	30 days	35 days
Diazinon	0·03	35 days	40 days
FOLIDOL E 605 forte	0·03	35 days	35 days

In this test, batches of 50 infested olives of both varieties were picked from the tree top and from the bottom outer branches for the evaluation of each product, in other words 4 × 50 olives were examined each time for each product. As the results in Table 4 show, the classification of the residual effect for the different products is the same as in test No. 6 (Table 3). LEBAYCID produced the longest residual effect also in this test.

In connection with the results of Test No. 5 (Palea Rumata), brief reference should be made to the relations between the residual effect of the product and the fruit weight and oil content of the olives, for apart from the spray concentration and potency of the product, the residual action may also be influenced by the size and the stage of growth of the olives. Therefore, in several tests, parallel to the evaluations of the residual action, the fruit weight and oil content of the olives were checked* at intervals of 0, 10, 20, 30, 40 and 55 days after spraying. The most interesting results obtained are those showing the differences between the olive varieties "Tsunati" and "Lianolia":

I. Olive variety "Tsunati"

Test No.	1	5
Fruit weight	1·39 gr	1·31 gr
Oil content in fresh substance	22·3 %	19·8 %
Oil content in dry substance	48·8 %	44·2 %
Quantity of oil in gr per single fruit	0·31 gr	0·26 gr

II. Olive variety "Lianolia"

Test No.	6	5
Fruit weight	0·79 gr	0·66 gr
Oil content in fresh substance	20·5 %	16·7 %
Oil content in dry substance	46·8 %	44·9 %
Quantity of oil in gr per single fruit	0·16 gr	0·11 gr

The fruit weight of the Lianolia variety is only about half that of the Tsunati variety, also the oil quantity in gr/fruit, which is calculated from the percentage oil content (in the fresh substance). As the fruit weight and the oil quantity in gr/fruit in Test No. 5 (Palea Rumata) are reduced by almost exactly a half for Lianolia, the residual effect ought to be approximately the same for both varieties. It should, however, be borne in mind, that is when the relations for the cube and for the sphere are considered, that the surface area generally drops from 100 % to 63 % when the volume is reduced from 100 % to 50 % so that, for example, the surface area of the small fruit undergoes a relative increase of approximately 25 %. The fruits of the Lianolia variety (with the smaller fruit weight), therefore, absorb relatively more spray solution per fruit than the fruits of the Tsunati variety; this is evident from the immersion tests and the calculations of Ayoutantis et al (7), as the following graph shows.

Accordingly, the residual effect ought to last longer on the small fruit varieties than on the large fruit varieties, which is partly the case (as shown by Table 4). In the official spraying recommendations, lower spray concentrations are, therefore, recommended for treatment of small fruit varieties than for large fruit varieties, also in view of the residue values.

* The oil analyses were kindly carried out by the Phytopathological Institute at Benaki (Kiphissia, Athens).

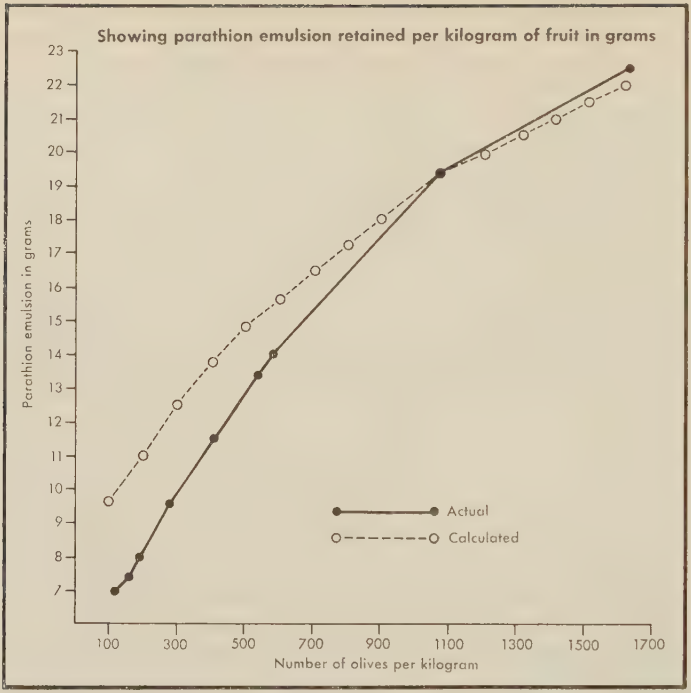

Fig. 8: Relationship between size of olive fruit and retained spray solution (according to Ayoutantis et al [7])

Residual effect for delayed new infestation

Tests 1 to 4 (Perivolia) were carried out in the first half of September, at a time when the *Dacus* population had been greatly decimated by the heat and the large abundance of parasites made it difficult to carry out a satisfactory evaluation. In these experiments, importance was attached to testing the effect on larvae of sprays carried out several weeks before the re-infestation which, under these circumstances, was anticipated in October. In fact, re-infestation occurred about October 10, in other words 30 to 40 days after spraying, and at a time when the experiments to test the effect on adult flies also began; these will be discussed later.

The evaluation results obtained for the residual effect that is produced against delayed new infestation, are given in Tables 5, 6 and 7. The tabulated figures include not only the numbers of living stages counted in each evaluation but also the dead larvae and the number of punctures and eggs that were found. It is evident from the data given in Tables 5 and 6 for the first evaluation (47 resp. 40 days after the spray application) that the number of eggs and punctures is of the same order of magnitude for all products; on the other hand, no dead larvae were found for DIPTEREX (0·1 and 0·15 %), FOLIDOL M 50 (0·05 %), FOLIDOL E 605 forte (0·05 %) and the untreated check,

Table 5: Test No. 1 and 2 (Perivolia). Residual effect for delayed new infestation

Spraying on			Sampling: 18. 10. (47 days) Examination: 20. 10. (49 days)												27. 10. (56 days) 30. 10. (59 days)													10. 11. (70 days) 12. 11. (72 days)						
			Dead							Living					Dead					Living							Living						
Test No. 1: 1. 9. 1958			No.				L_1		L_2	L_3	L_1		L_2	L_3	No.				L_1		L_2	L_3	L_1		L_2	L_3	S	No.			L_1		L_2	L_3	S
Test No. 2: 2. 9. 1958	Active ingredient %		olives												olives													olives						
			examined	infested	punctures	eggs	mM	oM			mM	oM			examined	infested	punctures	eggs	mM	oM			mM	oM				examined	infested	punctures	mM	oM			
Test No. 1																																			
DIPTEREX Emulsion	0·1	top*	25	25	49	15	—	—	—	—	10	11	2	1	50	42	74	15	7	-	6	3	15	-	9	3	12								
		bottom													50	43	87	16	8	-	4		17	4	9	3	3								
DIPTEREX Emulsion	0·15	top	25	25	40	14	—	—	—	—	8	7	5	2	50	31	54	12	—		4	1	16	1	5	4	2								
		bottom													50	20	28	11	2	—	-		6	2	2	3	1								
LEBAYCID Emulsion	0·06	top	25	25	31	10	9	7	—	—	3	2	—	—	50	46	98	17	7	7	1		22	11	—	—									
		bottom													50	33	47	15	1	2	3		13	3	—	1	—	50	50	104	6	—	4	14	13
LEBAYCID Emulsion	0·1	top	25	25	28	12	5	9	—	—	1	—	-	—	50	42	76	21	8	9	1	—	11	9	1	2	1								
		bottom													50	42	69	21	22	15	—	3	9	7	—	1	1	50	50	165	2	—	7	12	9
LEBAYCID Wettable Powder	0·06	top	25	25	34	16	5	7	—	—	1	—	—	—	50	42	79	24	11	10		1	18	3	1	—	—								
		bottom													50	22	37	12	6	4	1		8	2	—	1	1	50	50	148	5	—	5	11	2
LEBAYCID Wettable Powder	0·1	top	25	25	30	15	5	10	—	—	—	—	—	—	50	35	50	23	10	5	1	—	8	3	—	—	—								
		bottom													50	19	22	9	5	2	2	—	7	1	—	—		50	50	124	8	1	7	4	1
FOLIDOL M 50	0·05	top	25	25	29	5	4	—	—	—	12	5	2	—	50	50	97	10	4		1	—	30	1	21	3	2								
		bottom													50	36	46	4	—	-	1	2	24	—	9	5	4								
FOLIDOL E 605 forte	0·05	top	25	25	39	6	4	3	2	—	8	9	1	1	50	36	53	5	2	—	-		24	2	7	6	1								
		bottom													50	35	42	2	—	4	1	—	23	—	3		2								
4961	0·06	top	25	25	33	11	9	1	—	1	4	7	—	—	50	47	68	9	3	1	1	—	26	1	7	5	3								
		bottom													50	34	46	7	4	—	—	—	14	—	7	5	—								
4891 a	0·06	top	25	25	37	14	3	—	1	—	7	9	-	—	50	47	94	3	2	3	2	—	30	3	9	7	—								
		bottom													50	41	90	5	—	—	—	—	23	—	5	2	—								
Diazinon	0·06	top	25	25	31	12	3	5	—	—	3	4	—	—	50	32	30	11	4	3	—	—	24	2	1	—	—								
		bottom													50	30	39	14	1	1	1	—	20	1	-	—	—	50	50	137	14	—	12	15	9
Test No. 2																																			
LEBAYCID Emulsion	0·1	top													50	43	95	—	42	20	3	—	4	—	3	—	—								
		bottom													50	36	104	—	30	39	—	—	—	—	—	—	—	50	50	173	7	—	13	18	9
LEBAYCID Emulsion	0·15	top	25	25	35	7	6	19	—	—	—	—	—	—	50	34	51	9	19	4	2	—	10	6	—	—									
		bottom													50	42	65	6	18	8	1	—	6	—	—	—		50	50	130	2	—	16	25	14
Untreated	—	top	25	25	28	13	—	—	—	—	7	6	2	1	50	32	67	3	—	2	—	—	21	—	12	11	4								
		bottom													50	33	61	2	—	—	—	—	19	—	11	13	—								

* top = olives sampled from tree top
bottom = olives from bottom outer branches
mM = with galleries
oM = less galleries

whereas live larvae of all stages were present. It is concluded from these results that the insecticidal effect of these products had already died out by the time re-infestation occurred thirty days after the spray application. Better results were obtained for diazinon (0·06 %), although this product is not persistent for more than 40 days according to Tables 5 and 6. An exceptional product in this respect is LEBAYCID which was sprayed in these tests (Tables 5, 6, 7) at concentrations of 0·06 %, 0·1 %, 0·12 % and 0·15 % active ingredient. These concentrations were found to kill a high percentage of emerging larvae before they are able to tunnel in the fruit, even 40 days after the spray application; this is evident from the high mortality figures recorded for LEBAYCID in the column headed "L_1 less galleries" in Tables 5, 6 and 7. The residual effect is extended by increasing the concentration.

Fig. 9: In the trackless hilly districts, water and sprayers required for olive fruit fly control operations have to transported by pack-animals, which considerably increases the costs of spraying. Therefore, in order to counter shortage of water and to reduce costs for labour, dusters are used in some places

Average residual effect obtained

The durations of residual effect tabulated on P. 79, are derived from the results obtained in this test series; these figures must, however, be treated with certain reservation in view of the fact that tests were conducted for only a short period in autumn. Furthermore, these values apply only to autumn treatment of the olive varieties "Tsunati" and "Lianolia".

Using the same active ingredient concentration, LEBAYCID accordingly has an effect superior to that of the other products (Diazinon, FOLIDOL E 605 forte and FOLIDOL M 50). Furthermore, the residual effect of LEBAYCID is

Table 6: Test No. 3 (Perivolia). Residual effect for delayed new infestation

Spraying on 8. 9. 1958	Active ingredient %		Sampling: 18. 10. (40 days) Examination: 20. 10. (42 days) No. olives examined	infest.	punctures	eggs	Dead L_1 mM	Dead L_1 oM	Dead L_2	Dead L_3	Living L_1 mM	Living L_1 oM	Living L_2	Living L_3
DIPTEREX Emulsion	0·1	top*	25	25	36	7	—	—	–		17	3	4	3
		bottom												
DIPTEREX Emulsion	0·15	top	25	25	34	11	—	—	–	–	9	1	–	1
		bottom												
LEBAYCID Emulsion	0·06	top*	25	25	28	11	7	4	1		3	2	–	–
		bottom												
LEBAYCID Emulsion	0·1	top	25	25	34	14	5	6	—	–	4	1	–	–
		bottom												
FOLIDOL M 50	0·05	top	25	25	35	7	3	—	1		12	5	2	2
		bottom												
FOLIDOL E 605 forte	0·05	top	25	25	36	10	—	—	—	–	14	4	3	
		bottom												
Diazinon	0·06	top	25	25	30	12	—	—	2	—	7	1	–	
		bottom												
Untreated	—	top	25	25	28	5	–	—		–	12	3	5	3
		bottom												

Spraying on 8. 9. 1958	27. 10. (49 days) 4. 11. (57 days) No. olives examined	infest.	punctures	Dead L_1 mM	Dead L_1 oM	Dead L_2	Dead L_3	Living L_1 mM	Living L_1 oM	Living L_2	Living L_3	Living S
DIPTEREX Emulsion	50	42	75	—	—	—		28	1	15	7	4
	50	44	83	—	—		—	23	2	15	10	7
DIPTEREX Emulsion	50	14	16		—	—		8	1	2	1	—
	50	34	69	—	—			8	—	15	10	2
LEBAYCID Emulsion	50	37	48	3	22	–	–	13	5	—	—	—
	50	40	62	8	15	—	—	14	3		—	—
LEBAYCID Emulsion	50	42	55	1	40	—	—	—	- -	—	—	—
	50	39	66	4	40	—	—	4	—	—	—	—
FOLIDOL M 50	50	39	83	5	2	2		29	—	7	2	1
	50	37	55	3	2			21		7	5	—
FOLIDOL E 605 forte	50	28	30	—	2	—	—	25	—	16	7	3
	50	45	79	—	1	—	1	8		8	4	—
Diazinon	50	39	104	6	15	1	—	32	—	4	2	1
	50	27	46	1	1	2	—	12		8	5	1
Untreated	50	30	34	—	3		—	12	-	8	3	1
	50	19	31	2	—	—	—	9	1	2	4	1

Spraying on 8. 9. 1958	10. 11. (63 days) 13. 11. (66 days) No. olives examined	infest.	punctures	Living L_1 mM	Living L_1 oM	Living L_2	Living L_3	Living S
DIPTEREX Emulsion								
DIPTEREX Emulsion								
LEBAYCID Emulsion	50	50	158	—	—	14	9	10
LEBAYCID Emulsion	50	50	125	—	—	4	4	3
FOLIDOL M 50								
FOLIDOL E 605 forte								
Diazinon	50	50	109	10	—	15	12	17
Untreated								

* top = olives sampled from tree top
bottom = olives from bottom outer branches
mM = with galleries
oM = less galleries

Table 7: Test No. 4 (Perivolia). Residual effect for delayed new infestation

Spraying on 9. 9. 1958	Active ingredient %		Sampling: 27. 10. 1958 (48 days) Examination: 5. 11. 1958 (57 days)												10. 11. (62 days) 12. 11. (64 days)							
			Dead							Living					Living							
			No. olives examined	No. olives infested	No. punctures	L_1 mM	L_1 oM	L_2	L_3	L_1 mM	L_1 oM	L_2	L_3	S	No. olives examined	No. olives infested	No. punctures	L_1 mM	L_1 oM	L_2	L_3	S
LEBAYCID-Emulsion	0·06	top*	50	41	114	18	10	—	—	20	—	12	4	1								
		bottom	50	50	163	22	40	—	—	—	—	—	2	1	50	49	112	—	—	4	8	8
LEBAYCID-Emulsion	0·12	top	50	40	76	22	23	—	—	10	5	—	—	—								
		bottom	50	49	91	25	54	—	—	2	1	—	—	—	50	50	109	—	—	2	4	4
LEBAYCID-Wettable Powder	0·06	top	50	10	12	3	6	—	—	1	—	—	—	—								
		bottom	50	50	95	15	14	4	—	16	2	10	—	2	50	50	103	—	—	4	7	18
LEBAYCID-Wettable Powder	0·12	top	50	37	69	20	30	2	—	2	—		—	—								
		bottom	50	39	64	25	26	—	—	8	—	—	—	—	50	50	160	—	—	6	4	14
DIPTEREX-Emulsion	0·12	top	50	17	28	1	—	—	—	10	4	3	5	2								
		bottom	50	12	21	4	2	—	—	6	1	3	4	5								
4891a	0·06	top	50	44	86	10	6	—	—	38	6	4	2	3								
		bottom	50	39	62	16	6	—	—	20	14	1	5	7								
FOLIDOL E 605 forte	0·06	top	50	22	29	1	2	2	—	12	4	4	—	—								
		bottom	50	44	67	4	3	1	—	22	—	13	6	1								
FOLIDOL M 50	0·06	top	50	34	92	3	1	—	—	23	1	4	—	—								
		bottom	50	30	51	—	—	—	—	16	—	11	3	2								
LEBAYCID-Emulsion	0·1	top	50	50	105	8	60	4	1	4	—	—	—	—								
		bottom	50	47	84	4	61	2	1	2	1	1	—	—								
Untreated	—	top	50	48	148	1	—	1	—	12	—	12	19	5								
		bottom	50	50	181	—	—	1	1	9	—	11	10	21								

* top = olives sampled from tree top
bottom = olives from bottom outer branches

mM = with galleries
oM = less galleries

Concentration active ingredient	LEBAYCID	Residual effect in days Diazinon	FOLIDOL E 605 forte	FOLIDOL M 50
0·015 %	35—40			
0·03 %	40—45	35—40	25—30	25—30
0·06 %	45—50	40—45		
0·1 %	48—53			
0·12 %	50—55			
0·15 %	53—57			

extended by about 5 days when the concentration of active ingredient is doubled; this also applies to Diazinon. FOLIDOL E 605 forte and FOLIDOL M 50 have the same residual effect (about 20 to 30 days at a concentration of 0·03 % active ingredient), whereas the initial effect of FOLIDOL M 50 is better than that of FOLIDOL E 605 forte.

The other products (DIPTEREX, METASYSTOX, Thiometon, 4961, 4891a) cannot be considered for this use because of their unsatisfactory initial action and/or residual effect.

Effect against adult flies

In recent years, extensive tests have been carried out in Morocco and in Greece (4), in which preventive cover sprays, using organophosphorus compounds, were applied in early summer, especially on table olives, with the

Fig. 10: The trap sheets — in these tests a count was made every morning and evening of killed stages — were guarded by a night-watchman to ensure that there would be no outside interference with the tests also during the night

object of eradicating, inter alia, *Dacus oleae* adults and thus completely preventing puncturing of olives and, consequently, egg-laying. The quality of table olives is considerably lowered when they are punctured by olive fruit fly, for scars form at the punctured points on the green olives, appearing as blackish-brown spots. Therefore, the desired object of these sprays is to kill immediately all flies present on the crop, and also to eradicate, through the contact and stomach action of the residual deposit on the branches, leaves and fruits, any adults that later fly onto the trees or emerge from the ground, thus preventing oviposition. The concentrations of these sprays must be kept as low as possible because of the residue values.

Large-scale exact trap sheet experiments were conducted in 1957 by Orphanidis et al (28) to test initial action and residual effect of different products (0·05 % active ingredient) against imagines. These tests were carried out in autumn and not in early summer because it is most desirable, also in plantations of olives grown for yielding oil, that products sprayed to control larvae give a simultaneous effect against adult flies. When the residual effect of products against imagines is tested in late summer and autumn spray applications, it must be taken into consideration that newly emerging flies come from pupae located in the fruits themselves. As all products hitherto available are ineffective against pupae in the fruit, it is important to know whether, and how many days after the spray application, the newly emerging flies are killed by the spray deposit on fruits and leaves.

In our trap sheet experiments which were conducted from mid-October until early November, LEBAYCID was tested together with a comparison product*. Consideration was given to the tesults of Ophanidis et al (28) in selecting the comparison product. The sprays were applied at concentrations of 0·03 and 0·015 % active ingredient. To test the initial effect, two trees each were sprayed on October 12, 1958 (trap sheet test No. 1) with LEBAYCID Emulsion and the comparison product; trap sheets measuring 60 square metres were placed under each tree, and the killed insects were counted at intervals of 1, 3, 5 and 10 hours after the application. The results in Table 8 show that LEBAYCID gives a satisfactory initial effect. In order to test the residual effect, 12 square metre trap sheets were laid out under each tree from the third day of this test, and mortality was registered until the 21st day. Although this test was affected by wind and rain, flies killed by both products were still found on the 20th day of the test. The trees were sprayed again, however, on the evening of the 21st day, and on the next day a large number of dead flies were found under these trees.

As will be seen from Table 9, trap sheet test No. 3 was carried out at about 5 p. m. on October 27, 1958, with three replications. These sprays were applied in a mixed orchard containing 120 olive, 220 mandarin and 30 orange trees. All these trees (olives, mandarins and oranges) had already been sprayed with LEBAYCID Emulsion (0·03 % active ingredient) on October 12, 1958, 16 days beforehand. It should also be mentioned that trap sheet test No. 1 had been carried out in the same orchard on Oct. 12, 1958; the good results obtained with LEBAYCID in this test are given in Table 8. The results in Table 9 show that for the spray application on Oct. 27, 1958, the new spray applied 16 days

* N-monomethylamide of 0,0-dimethyldithiophosphoryl acetic acid.

Table 8: Trap sheet test No. 1 (Chania). Mortality obtained in days after spray application

Spraying on Oct. 12, 1958 at 8 a.m.		12. 10.					13. 10.	14. 10.	15. 10.	16. 10.	17. 10.	18. 10.	19. 10.	20. 10.	21. 10.	22. 10.	23. 10.	24. 10.	25. 10.	26. 10.	27. 10.	28. 10.	29. 10.	30. 10.	31. 10.	1. 11.	Sum for all days	in % in relation to LEBAYCID		2. 11.	
Interval after spraying	Tree No.	1 hr.	3 hrs.	5 hrs.	10 hrs.	Sum 1st day	2nd day	3rd day*	4th day	5th day	6th day	7th day	8th day	9th day	10th day	11th day	12th day	13th day	14th day	15th day	16th day	17th day	18th day	19th day	20th day	21st day				1st day	in % in relation to LEBAYCID
								wind										rain												
LEBAYCID Emulsion 0·03 % active ingredient	1	146	77	8	9	240	—	3	—	—	—	—	5	25	13	21	6	—	—	—	6	23	12	7	4	0	365			370	
	2	183	65	9	17	273	—	1	—	—	—	—	10	22	10	7	13	—	—		3	35	21	10	6	9	420		new spray application 2. 11.	340	
	Σ	329	142	17	26	513	—	4	—	—	—	—	15	47	23	28	19	—	—	—	9	58	33	17	10	9	785			710	
	∅	165	71	8	13	257																					392	100		355	100
N-monomethylamide of 0·0-dimethyldithio-phosphoryl acetic acid 0·03 % active ingredient	1	91	50	6	4	151	—	2	—	—	—	—	2	5	4	9	4	—	—	—	3	18	1	4	8	1	212			131	
	2	140	36	6	3	185	—	1	—	—	—	—	9	18	7	4	16	—	—		4	20	11	4	3	1	282			175	
	Σ	231	86	12	7	336	—	3	—	—	—	—	11	23	11	13	20	—	—	—	7	38	12	8	11	2	495			306	
	∅	116	43	6	3	168																					247	63		153	43

* On 1st day, 60 sq. m. trap sheets were laid out (only during the day);
from 3rd—16th day, 12 sq. m. sheets (day and night)
from 17th day, 30 sq. m. sheets, following new spray application on Nov. 2, 60 sq. m. sheets were used

after the first one, led to a high rate of mortality equal to that given by the first spray. This trap sheet test was evaluated for mortality for a period of 10 days, using 60 square metre trap sheets per tree.

Considerable mortality was registered on each of the 10 days for both products; LEBAYCID gave better results than the comparison product. Another spray application was carried out on the evening of November 1, 1958, again using 0·03 % active ingredient and 60 square metre trap sheets per tree. This time mortality was registered for 5 days, as shown in Table 9.

It is also noted from Table 9 that in trap sheet tests No. 3 and 4, a third spray was carried out on the evening of Nov. 6, 1958, this time using only 0·015 % active ingredient, ten and five days, respectively, after the second spray (and 16 and 21 days, respectively, after the first spray). The figures for the count on Nov. 7, 1958 show that mortality was very high for all trees, whether last sprayed 10 or 5 days beforehand; LEBAYCID gave a bigger kill than the comparison product. From the results of these tests, it can be concluded that LEBAYCID has a most considerable effect against adults. It is, however, doubtful whether this can be interpreted as a residual effect in the sense that over a long period after the spray application, all newly migrating flies attacking the trees and new flies emerging from the fruits will be killed by the contact and stomach action of the spray deposit on the fruits and leaves, before they are able to puncture the fruits and lay eggs (this is especially doubtful in view of the fact that the concentrations cannot be appreciably increased because of the residue values, for such increase would be necessary to improve the contact action of the spray deposit).

The figures tabulated below show that LEBAYCID gave a good effect in all tests.

	Dead flies/tree in observation period			
Trap sheet test No.	1	3	4	5
Conc. of active ingredient	0·03 %	0·03 %	0·03 %	0·015 %
Observation period	21 days	10 days	5 days	1 day
LEBAYCID	392 (100)	726 (100)	630 (100)	289 (100)
N-monomethylamide of 0,0-dimethyldithiophosphoryl acetic acid	247 (63)	597 (82)	358 (57)	175 (61)

III. Summary of test results

Trial controls of olive fruit fly were carried out in the area around Chania in Crete, from September 1 to November 16, 1958. The initial action and residual effect (against larvae and adults) of LEBAYCID and several other test products were tested in comparison with diazinon and E 605 forte. Furthermore, FOLIDOL M 50 was tested in comparison with FOLIDOL E 605 forte because FOLIDOL M 50 is less toxic. As Thiometon had also been used in recent years against olive fruit fly, METASYSTOX was included in the tests for the purpose of comparison. The results obtained in these tests permit the following conclusions to be drawn:

Table 9: Trap sheet tests No. 3, 4, and 5 (Chanis). Mortality obtained in days after spray application

Spraying on October 27, 1958 at 5 p. m.		1st day 28. 10.			2nd day 29. 10.			3rd day 30. 10.			4th day 31. 10.			5th day 1. 11.			6th day 2. 11.			7th day 3. 11.			8th day 4. 11.			9th day 5. 11.			10th day 6. 11.			Sum for all days			in % in relation to LEBAYCID			1st day 2. 11.			in % in relation to LEBAYCID
Counts	Tree No.	7 a.m.	5 p.m.	daily sum	7 a.m.	5 p.m.	daily sum	7 a.m.	5 p.m.	daily sum	7 a.m.	5 p.m.	daily sum	7 a.m.	5 p.m.	daily sum	7 a.m.	5 p.m.	daily sum	7 a.m.	5 p.m.	daily sum	7 a.m.	5 p.m.	daily sum	7 a.m.	5 p.m.	daily sum	7 a.m.	5 p.m.	daily sum	7 a.m.	5 p.m.	daily sum				7 a.m.	5 p.m.	daily sum	
Test No. 3	1	82	51	133	19	10	29	2	2	4	4	8	12	12	16	28	—	—	—	—	—	—	—	—	—	—	—	—	—	Rain	—	—	14	14	303	286	589						
LEBAYCID	2	92	76	168	38	91	129	22	10	32	12	12	24	11	26	37	41	11	52	32	9	41	34	14	48	21	23	44			14	14	303	286	589		New application 6. 11. 5 p.m.	*Test No. 5*					
Emulsion	3	137	89	226	36	184	220	32	13	45	10	27	37	25	46	71	34	40	74	38	19	57	18	21	39	27	50	77			16	16	357	505	862			LEBAYCID	210	38	248	
0·03 %	Σ	311	216	527	93	285	378	56	25	81	26	47	73	48	88	136	75	51	126	70	28	98	52	35	87	48	73	121	—	30	30	660	791	1451	100		Emulsion	255	43	298	
	Ø	104	72	176	31	95	126	19	8	27	8	16	24	16	29	45	38	25	63	35	14	49	26	18	44	24	37	61	—	15	15	330	396	726			0·015 %	465	81	546	100
																																						233	40	273	
Comparison	1	41	37	78	16	18	34	15	3	18	11	6	17	15	14	29	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—			Comparison				
product*	2	83	53	136	36	123	159	13	6	19	9	14	23	11	11	22	26	27	53	29	4	33	19	9	28	12	28	40	Rain	11	11	238	286	524			product*)	129	16	145	
0·03 %	3	103	82	185	32	141	173	7	6	13	10	23	33	20	26	46	50	20	70	33	18	51	27	16	43	19	25	44		13	13	301	370	671			0·015 %	166	54	220	
	Σ	227	172	399	84	282	366	35	15	50	30	43	73	46	51	97	76	47	123	62	22	84	46	25	71	31	53	84	—	24	24	539	656	1195	82			295	70	365	67
	Ø	76	57	133	28	94	122	12	5	17	10	14	24	15	17	32	38	24	62	31	11	42	23	13	36	15	27	42	—	12	12	270	328	597				148	35	183	
Check	1	—	—	—	1	—	1	-	—	-	—	—	—	—	—	—	—	—	—					-	—	1		1	—	—	—	2	—	2			Check	—	—	—	

Spraying on 1. 11. 5 p.m.		1st day 2. 11.			2nd day 3. 11.			3rd day 4. 11.			4th day 5. 11.			5th day 6. 11.			Sum for all days			in % in relation to LEBAYCID			1st day 7. 11.			in % in relation to LEBAYCID
Counts		7 a.m.	5 p.m.	daily sum	7 a.m.	5 p.m.	daily sum	7 a.m.	5 p.m.	daily sum	7 a.m.	5 p.m.	daily sum	7 a.m.	5 p.m.	daily sum	7 a.m.	5 p.m.	daily sum				7 a.m.	5 p.m.	daily sum	
Test No. 4																										
LEBAYCID	1	265	105	370	34	43	77	43	26	69	35	30	65	Rain	48	48	377	252	629							
Emulsion	2	250	90	340	29	53	82	60	29	89	38	41	79		40	40	377	253	630			LEBAYCID	202	50	252	
0·03 %	Σ	515	195	710	63	96	159	103	55	158	73	71	144	—	88	88	754	505	1259		New application 6. 11. 5 p.m.	Emulsion	244	115	359	
	Ø	258	97	355	32	48	80	52	27	79	37	35	72	—	44	44	377	253	630			0·015 %	446	165	611	100
Comparison	1	110	21	131	24	21	45	27	14	41	25	27	52	Rain	30	30	186	113	299				223	83	306	
product*	2	122	53	175	29	40	69	79	24	103	27	22	49		21	21	257	160	417			Comparison	93	24	117	
0·03 %	Σ	232	74	306	53	61	114	106	38	144	52	49	101	—	51	51	443	273	716	57		product*)	153	63	216	
	Ø	116	37	153	26	31	57	53	19	72	26	25	51	—	26	26	222	136	358			0·015 %	246	87	333	55
																							123	44	167	

Note: All mortality figures registered on 60 sq. m. trap sheet/tree

* N-monomethylamide of 0,0-dimethyldithiophosphoryl acetic acid

Effect against larvae

1. The initial effect given by the different products (using the same active ingredient concentration) against older larval stages varies most considerably. The best initial effect is produced by LEBAYCID, followed by 4961, FOLIDOL M 50 and DIPTEREX; practically 100 % of the older larval stages were killed at active ingredient concentrations of 0·03 % and 0·015 % after only 2 days. In contrast to FOLIDOL M 50, it took FOLIDOL E 605 forte five days to give a 100 % kill of L_2 and L_3. Five days after application of 4891 a, diazinon, Thiometon and METASYSTOX, a considerable percentage of live L_2 and L_3 were still present on the trees. The initial effect against older larval stages is of less importance than the residual effect because timely sprayings should usually be carried out when new infestation is just starting, in other words at a time when only eggs and L_1 are present, and when the fruits are still unharmed.

   Growers cannot always keep to such a schedule, however, so that a good initial effect against olives is also of interest to growers when selecting a suitable product because the fruits, in which L_2 and L_3 are already tunneling at the time of spraying, usually recover from feeding damage and ripen as the result of the immediate eradication of these larvae.

2. The residual effect (or persistence, i. e. the period following the spray application, in which larvae hatching from newly laid eggs are killed by active ingredient still present in the fruit) given by the different products at the same concentration of active ingredient, also varies most appreciably.

   The most persistent product in these tests was LEBAYCID, followed by diazinon, FOLIDOL E 605 forte and FOLIDOL M 50. Thiometon and METASYSTOX are practically as persistent as the parathion products, but their initial effect is inadequate. 4891 a and 4961 are unsuitable because of their very short residual effect. The least persistent of the tested products was DIPTEREX.

   On calculating the averages for the test results, the following persistence values are obtained for LEBAYCID in comparison with diazinon, FOLIDOL E 605 forte and FOLIDOL M 50, at the different concentrations tested:

Active ingredient concentration	Persistence expressed in days			
	LEBAYCID	Diazinon	FOLIDOL E 605 forte	FOLIDOL M 50
0·015 %	35—40			
0·03 %	40—45	35—40	25—30	25—30
0·06 %	45—50	40—45		
0·12 %	50—55			

The residual effect of LEBAYCID is accordingly lengthened by approximately 5 days upon doubling the concentration of active ingredient; this is also about the same for diazinon. When applied at the same concentration of active ingredient, LEBAYCID is superior in its residual effect to the other products. FOLIDOL E 605 forte and FOLIDOL M 50, applied at an active

ingredient concentration of 0·03 %, gave a residual effect of 25 to 30 days in our tests, so that on the basis of the various test data we recorded, these two products can be rated as being of equal persistence.

It should again be mentioned that these results were obtained on the olive varietes "Tsunati" and "Lianolia". The oil content of the fruits ranged from 15 to 25 %; the fruit weight for the "Lianolia" olive variety was 0·5 to 1 gr, and for the "Tsunati" variety between 1 and 1·5 gr.

Effect against flies

Products capable of controlling adult flies are especially important in table olive growing. Eradication of adult flies before they lay their eggs prevents puncturing of table olives. The quality of table olives is appreciably lowered once the fruits are pierced by the olive fruit fly because the punctures develop into scars on the green olives, forming dark spots which render the fruit unfit for consumption. Extermination of adult flies present on the crop at the time of spraying is also desirable in plantations of olives grown for yielding oil.

A special problem in sprays against adult flies is the residual effect. New infestations that occur after spray applications, are caused in spring by flies emerging from pupae in the soil, and in summer and autumn by flies that emerge from pupae in the fruits. All products so far tested are ineffective against pupae in the fruit. New flies migrating to the crop and emerging from the fruits after the spray application, therefore, have to be eradicated by the contact and stomach action of the residual spray deposit on the branches, leaves and fruits. Because of regulations governing residues of pesticides in crops, spray concentrations have to be kept as low as possible, except for spray applications in spring.

In our trap sheet tests, attention was given to this problem by comparing LEBAYCID with N-monomethylamide of 0,0-dimethyldithiophosphoryl acetic acid at concentrations of 0·03 and 0·015 % active ingredient. LEBAYCID gave a bigger kill than the comparison product in these tests. Mortality was still registered 20 days after spraying. Repeated spray applications carried out 20, 10 and 5 days after the first spray application, gave an equal degree of mortality. A residual effect in the sense of killing new batches of flies migrating to the trees and emerging from the fruits before they can lay their eggs, whereby such eradication continues for a lengthy period after the spray application and is effected by the contact and stomach action of the spray deposit on the leaves and fruits, can therefore only be brought about by large-scale spray applications.

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Trial Controls of Olive Fruit Fly — Field Tests in Corfu

by M. Boness

The introduction of cover sprays with phosphoric esters has for the first time rendered possible full control of olive fruit fly *(Dacus oleae)*, regarded as being the insect pest of greatest economic importance in the Mediterranean zone.

Products at present used in the field give an almost 100 % control when they are properly applied, but nevertheless they still leave much to be desired; apart from toxicological questions and residue problems that require solving, there is especially a need for improved economy.

The great majority of all olive crops bring in only a very slight profit, and very many trees grow on land of a nature which presents very serious difficulties to the technical implementation of control operations and the cartage of water. Therefore, a product is urgently needed which at the lowest possible dosage and in 1, or at the most 2 applications will prevent damage to the crop during the whole of the main infestation period. This main period of infestation usually extends, despite large local and regional deviations, from the end of the summer lull in infestation — caused by the heat — (from the end of August to mid-September) until the fruit begins to ripen (between mid-October and late November), on oil-yielding olives cultivated in Greece.

LEBAYCID® had already proved in laboratory and field trials to have a most persistent action against eggs and larval stages of several *Trypetidae*, including olive fruit fly, in fruits of many varieties, and therefore, also in view of its favourable toxicological values, this product appeared to hold much promise of success also for the use under discussion.

® = Registered trade-mark of Farbenfabriken Bayer Aktiengesellschaft, Leverkusen.

In addition to Crete*, the island of Corfu was also chosen for carrying out the tests. This island has the highest amount of rainfall — 1100 millimetres and more — in the whole of the Greek territory (apart from the Epirus Mts.), and high relative humidity is often recorded also in summer. As a result, regular occurrence of *Dacus* is expected on at least a fair percentage of the roughly 3 million olive trees growing on this island.

A disadvantage proved to be the fact that the mass of olive trees, growing in relatively dense plantations, are 300 to 400 years old, whereas compact young orchards are largely wanting. In the few plantations on the plain, it is practically impossible to carry out exact tests when growth is vigorous (at a tree height of approx 15 metres, and above), whereas in the rocky hill country exposure, fruit set, degree of maturity and infestation are factors that may considerably vary from one tree to another.

In order to obtain useful results nevertheless, the evaluation of the tests was carried out at intervals of one week, starting 4 to 5 days after the treatment. Samples from different parts of all treated trees, which depending upon infestation, included 50 to 100 and in extreme cases up to 200 olives, were examined under a binocular within 24 hours for infestation by living eggs, larvae and pupae.

This examination provided information on both the initial effect against all stages and the residual effect; an indication that the latter is fading is given by the occurrence of young larvae.

Most of the spray applications were carried out on hill slopes near to Benitsa, about 100 metres a. s. l. on the east coast of the island; a portable motor-powered sprayer of type "Platz Patria Z" was used. The 1st spray was applied on August 27, 1958, the 2nd on September 1, the 3rd on September 8, and the 4th on October 3, 1958. A 5th and a 6th spray application were carried out on September 10 and September 15, 1958, respectively, in the interior of the island near Ropa on a dry hilly ridge about 100 metres a. s. l.; here, a hand-operated, high-pressure cart-mounted sprayer was used. Other tests conducted could not be evaluated due to a lack of infestation.

The following products were applied at the given strengths:

LEBAYCID Emulsion	0·1 %	active material in test 1 and 5
LEBAYCID Emulsion	0·06 %	active material in test 1, 2, 3, 4, 5, 6
LEBAYCID Wettable Powder	0·06 %	active material in test 2
4961	0·06 %	active material in test 3 and 4
4891 a	0·06 %	active material in test 1 and 5
Diazinon	0·06 %	active material in test 3
FOLIDOL® M 50	0·05 %	active material in test 1, 4, 5
E 605® forte	0·05 %	active material in test 1, 4, 5
DIPTEREX® Emulsion	0·15 %	active material in test 3

The volume of liquid applied per tree varied according to tree size and growth, and corresponded to conventional practice. It was not always possible to give the trees full coverage due to the dense foliage.

* A report on the tests in Crete is given on Page 52—89 of this edition.

The temperatures were in the region of 20 to 25 °C during the treatments, rising to almost 35 °C only on August 27. There was very heavy rainfall on September 2 and 3, and from October 20—22. The maximum daily temperature was in the region of 35 °C until September 2, and afterwards usually between 25 and 30 °C, except for a spell of cold weather after October 15.

Apart from an uneven rate of infestation, a high degree of mortality brought about by ichneumon fly attacks and the effect of the heat made it difficult to evaluate the test results during the first few weeks.

The initial effect after 1 and 2 weeks was, therefore, calculated for all tests according to the formula of Abbott, using the modification of Schneider-Orelli (based on % mortality).

Effect against larvae of all stages

	% active material	after 1 week	after 2 weeks
LEBAYCID Emulsion	0·1	100	100
LEBAYCID Emulsion	0·06	97	100
LEBAYCID Wettable Powder	0·06	100	100
4961	0·06	94	90
4891 a	0·06	5	22
E 605 forte	0·05	70	83
FOLIDOL M 50	0·05	87	80

In the case of E 605 forte and FOLIDOL M 50, a very high mortality rate in the corresponding control plots had an unfavourable influence on the recorded values. In the DIPTEREX and diazinon plots, the initial infestation was too weak for a reliable determination of efficacy to be made, although this was fairly high.

Occurrence of young larvae in the different tests

LEBAYCID Emulsion	0·1 % active material:	still no occurrence at end of test after 57 resp. 45 dys.
LEBAYCID Emulsion	0·06 % active material:	after 41, 35, 38, 45 dys. still none after 40 dys.
LEBAYCID Wettable Powder	0·06 % active material:	after 41 dys.
4961	0·06 % active material:	after 29, 20 dys.
4891 a	0·06 % active material:	after 16, 12 dys.
Diazinon Emulsion	0·06 % active material:	after 23 dys.
E 605 forte	0 05 % active material:	after 22, 20, 30 dys.
FOLIDOL M 50	0·05 % active material:	after 30 dys. still none after 20, 30 dys.
DIPTEREX Emulsion	0·15 % active material:	after 18 dys.

Application of LEBAYCID Emulsion at a dosage of 0·1 % active ingredient especially gave a kill of most eggs before they hatched. Neither the olive fruit fly pupae nor the ichneumon flies parasitizing the *Dacus* larvae were killed by any products. No phytotoxic effects were caused.

In order to establish the effect on *Dacus* imagines, trap sheets measuring 2 × 2 metres were spread out under 2 young trees (crown diameter of 1 to 2 metres), and on October 14 0·5 litre of a solution of LEBAYCID Emulsion (0·1 % active ingredient), containing an addition of 0·5 % liquid hydrolysate of yeast resp. maize protein, was applied to each tree.

Mortality recorded:	after 2 hrs.	7 flies
	1 dy.	40 flies
	2 dys.	130 flies
	3 dys.	6 flies
	4 dys.	12 flies

The test was abandoned on the 5th day because of rain and cold weather which curbed the flying activities of the insect. The variations in the values are due to the changeable weather that prevailed during the test.

Another trap sheet test run concurrently under large trees using 0·06 % active ingredient, produced a rate of mortality that decreased only slightly during the 6 days it was in progress. In addition, a large number of leaf-hoppers, ants, leaf weevils (mostly *Apion* species), pentatomids, and other *Diptera* species were found dead on the trap sheets.

Although these tests fail to give a full impression of the effect produced by LEBAYCID against *Dacus oleae* imagines, due to the weather disturbances that were experienced at the time, they do, however, show that the action of this product is most considerable, both with and without an addition of bait, and that the product deserves further testing especially for use on table olives.

Summary:

Trial controls of the olive fruit fly *(Dacus oleae)* were carried out under practical conditions in Corfu in autumn, 1958. LEBAYCID Emulsion, applied at dosages of 0·1 % and 0·06 % active ingredient, and LEBAYCID Wettable Powder, applied at 0·06 %, gave a 100 % kill of larvae colonizing the treated crop.

Both products completely prevented reinfestation for 5—6 weeks, when applied at 0·06 % active ingredient; a 0·1 % dosage produced a residual effect of at least 6 to 8 weeks.

4961 produced a good initial action but protected the crop against reinfestation for only 3 to 4 weeks. 4891 a is unsuitable for the control of olive fruit fly. FOLIDOL M 50 gave the same initial effect as E 605 forte, but its residual effect was found to be slightly better than that of the latter.